ANNUAL REPORT II:
Applied Research on Use of Native Plants for Coastal Wetland Restoration on O'ahu

May 2002 - May 2003

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To

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This final report covers work completed from May 2002 through May of 2003. This report also provides a summary of conclusions from outplanting field trials conducted at Pearl Harbor NWR and germination experiments conducted at the University of Hawai`i at Manoa. For work completed from May 2001 through May 2002 please review Appendix 2: Annual Report: June 2002. For further information and specifics on experimental design and monitoring protocol see Appendix 3: Research Proposal.

I. Research Overview

Seedlings of seven native wetland plant species were outplanted at Honouliuli Unit of Pearl Harbor NWR (hereafter referred to as Honouliuli) during the spring of 2002. Outplanting field trials were initiated to provide information on restoring native plant species to wetland habitat in Hawai`i. Little is known about restoring wetland habitat in Hawai`i with native plants. Information from this study was intended to increase knowledge on how to outplant native wetland plants; discern which plants are suited for various habitats; which native species compete best against alien weeds; long-term maintenance requirements; and potential use of native plants by native waterbirds as food, cover and nesting material.

Four sedge species, *Bolboschoenus maritimus* (L.) Palla (makai), *Cyperus javanicus* Houtt. (`ahu `awa), *Cyperus laevigatus* L. (makaloa), and *Cyperus polystachyos* Rottb., and three groundcover species, *Jacquemontia ovalifolia* (Choisy) (pa`uohi`iaka), *Sesuvium portulacastrum* (L.) L (`akulikuli), and *Sporobolus virginicus* (L.) Kunth (`aki`aki) were investigated during this study. Outplanting of native species occurred between March 16 and March 30, 2002. Monitoring of outplanted populations continued from April 2002 through April of 2003.

Two hundred and sixty *C. javanicus*, 300 *C. polystachyos,* 300 *S. portulacastrum*, and 380 *S. virginicus* seedlings were planted the week of March 16 – March 23, 2002. The following week, March 24 – March 31, 300 *C. laevigatus* seedlings and 260 *B. maritimus* transplants, culled from existing populations at Honouliuli, were outplanted. Due to nursery error, 300 *J. ovalifolia* seedlings were not ready for outplanting until early May 2002. This amounts to a total of 2,100 seedlings/transplants outplanted.

Seedlings were planted in single-species blocks with blocks of 1.25 m x 6.5 m for sedge species and 2 m x 3.5 m for groundcovers. There were ten blocks per species and, within each outplanting block, there were 6 permanent 0.25 m^2 plots (four planting plots and two control plots).

Outplanted seedlings were monitored twice monthly from April 1, 2002 until April 1, 2003. Parameters monitored, depending on species, included survival, percent cover, physical appearance, reproductive status, maximum height and number of shoots/tillers. Percent cover of non-planted species was also recorded twice monthly in each of the 420 permanent plots. Additional limited monitoring of survival, physical appearance and reproduction of the four sedge species was conducted in early May of 2003.

Soil salinity and soil temperature readings were taken in each plot twice a month from July 14, 2002 through April 1, 2003. Measurements of soil salinity and soil temperature, prior to July 14, 2002, were sporadic due to equipment failure. The Aquaterr Ec-200 meter used to measure soil parameters was sent back to the manufacturer for repairs in May 2002 and was not returned until July 2002. Measurements of soil moisture were taken in March 2002 and again from late-July of 2002 through October 2002. At this point the soil moisture readings became questionable, thus, soil moisture measurements ceased in November 2002.

Three separate germination experiments were conducted in the laboratory at the University of Hawai`i at Manoa beginning in August 2002 and ending in April 2003. These experiments were designed to test the germanability of the four sedge species, *B. maritimus, C. javanicus, C. laevigatus,* and *C. polystachyos*, under different conditions of salinity, moisture and temperature.

Monitoring of native bird use of outplanted species was initiated in April 2002. This observational study included a five-minute scan/observation period after every hour of plant monitoring / field work. This scan was intended to chart sightings of native birds in stands of native vegetation, duration in these stands and evidence of feeding, hiding in and/or using native plants for nesting material. However, due to lack of results, this work was suspended in July 2002. Personal communication with wetland managers, researchers, and biologists and literature reviews supplemented the observational study.

II. Results: Outplanting Field Trials

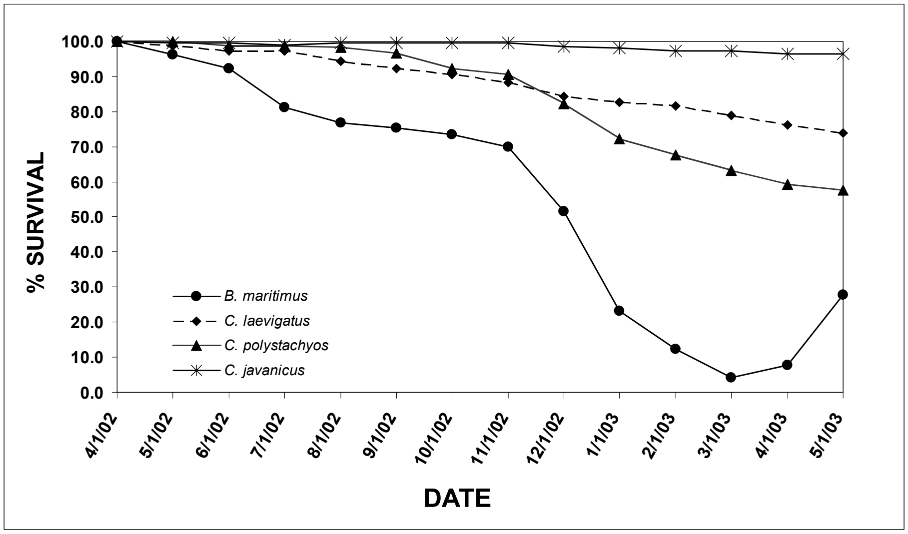
Survival and Physical Appearance

Outplanted seedlings of all species initially thrived in their new environment at Honouliuli. This was especially noticeable in seedlings of *C. javanicus, C. polystachyos*, and *S. virginicus* which, within two weeks of outplanting, appeared even healthier, stouter and richer in color than when received from the nursery.

This initial success was possibly in part due to mild and wet conditions that continued through May of 2002. Monthly rainfall recorded at Honouliuli for April and May of 2002 was 22.86 and 88.90 mm respectively. Additionally, to promote establishment of outplanted seedlings, each plot received 3.8 liters of supplemental water once a week for the month of April and 3.8 liters every other week during the month of May 2002. After May 2002 no additional water was supplied to the seedlings.

Despite drier weather, total rainfall for June through August 2002 was 17.78 mm, and lack of supplemental water, the vast majority of outplanted individuals survived the summer. More than 90% of outplanted individuals of all species (except *B. maritimus*) survived through September 2002 (Fig.1). No marked decline in survival, for any species, except *B. maritimus*, was noticeable until December 1, 2002 at which point several species dropped below 90% survival.

a)



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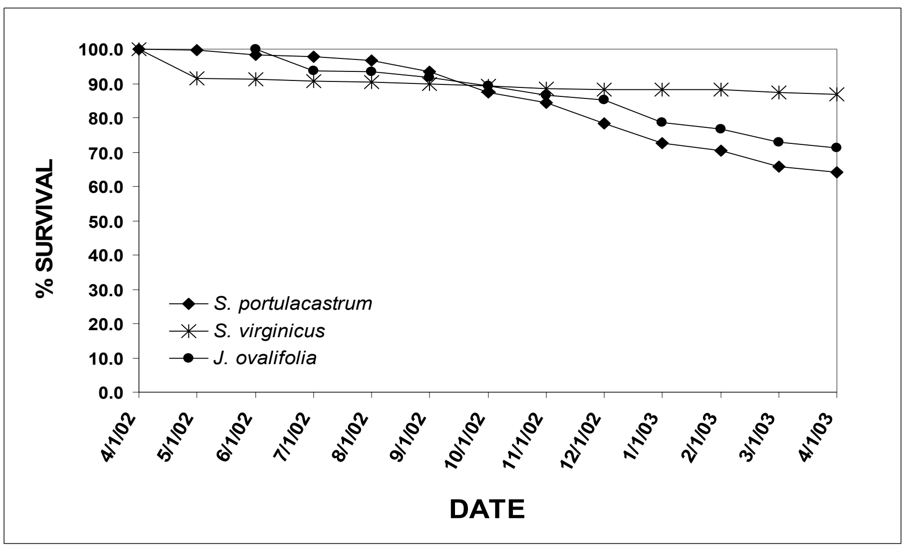


Figure 1. Monthly percent survival of (a) outplanted sedge species and (b) outplanted groundcover species April 1, 2002 to May 1, 2003

One year after outplanting 67.3%, of the original 2,100, plants were still alive (Table 1). *C. javanicus* had the highest survival rate (96.5%) with 251 of the original 260 plants still alive one year post-outplanting. Despite being one of the most common native species found growing naturally at Honouliuli, *B. maritimus* individuals demonstrated the lowest survival rate (7.7%) in April 2003, one year post-outplanting. However, this might represent just annual "die-back". By May 1, 2003 new shoots of *B. maritimus* were re-appearing next to outplanting marker tags. These new shoots, which presumably sprouted from underground tubers of the original transplanted individuals, increased *B. maritimus* percent survival to 27.7% by early May 2003 (Fig. 1).

Table 1. Quarterly survival rates of outplanted native wetland plants.

SPECIES	April 1, 2002 % Survival	August 1 2002 % Survival	December 1, 2002 % Survival	April 1, 2003 % Survival
Sedges				
Bolboschoenus maritimus (makai)	100%	76.9%	51.5%	7.7%
Cyperus laevigatus (makaloa)	100%	94.3%	84.3%	76.3%
Cyperus javanicus (`ahu `awa)	100%	99.6%	98.5%	96.5%
Cyperus polystachyos	100%	98.3%	82.3%	59.3%
Groundcovers				
Jacquemontia ovalifolia (pa`uohi`iaka)	100%	93.7%	85.3%	71.3%
Sesuvium portulacastrum (`akulikuli)	100%	97.7%	78.3%	64.0%
Sporobolus virginicus (`aki `aki)	100%	90.3%	88.2%	86.8%

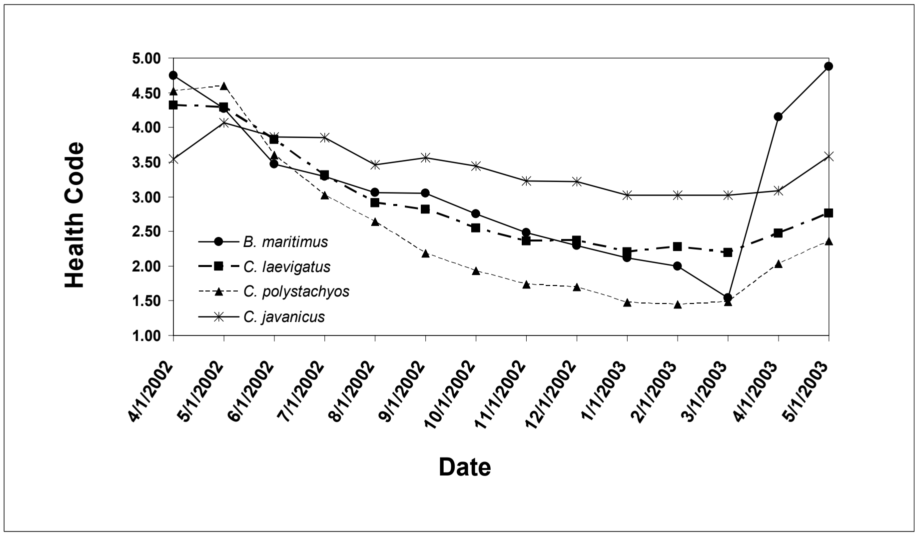
* numbers reflect totals on May 1st, 2002

Despite high overall survival rates, physical appearance of outplanted individuals started declining by June of 2002 (Fig. 2). Physical appearance, which was assessed by observing the percent of dark-green leaves and shoots versus the percent of yellowing or dead and dying leaf/shoot parts, was rated on a scale of 1 to 5 with 1 being very poor "health" (81-99% of individual dead or dying) and 5 being very healthy (less than 20% of individual dead or dying). These ratings will hereafter be referred to as "health-code" values.

The decline in physical appearance was most noticeable in *B. maritimus, C. laevigatus, C. polystachyos,* and *S. portulacastrum*. Individuals of *C. polystachyos* and *S. portulacastrum* exhibited the poorest physical appearance of any species in February 2003 with average health-codes of 1.45 ± 0.77 and 1.55 ± 0.87 respectively.

Physical appearance of *J. ovalifolia* individuals stayed relatively steady from June until August 2002, peaking with an average health-code of 4.67 on August 1, 2002. After August, physical appearance of *J. ovalifolia* steadily declined. Average physical appearance of *C. javanicus* individuals showed the least fluctuation throughout the study period with a maximum average health-code of 4.07 ± 1.25 on May 1, 2002 and a minimum average health-code of 2.97 ± 1.16. Interestingly, beginning in April 2003 the health of almost all species had started to rise. This was primarily due to the increase in production of new, healthy shoots/tillers by most of the study species which began in late March.

a)



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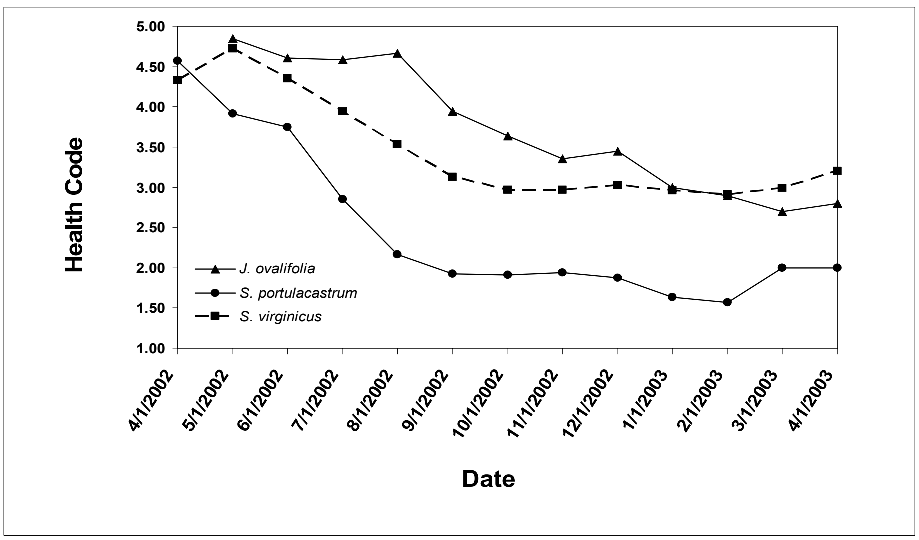


Figure 2. Physical appearance/health of (a) outplanted sedges and (b) outplanted groundcovers. Health-code: 5 = 0-20% dead or dying; 4 = 21-40% dead or dying; 3 = 41-60% dead or dying; 2 = 61-80% dead or dying; 1 = 81-99% dead or dying.

<u>Growth</u>

Vegetative growth of outplanted species was assessed by measuring the number of shoots per individual and/or the maximum height of each individual. Maximum height, measured for all species except *J. ovalifolia* and *S. portulacastrum,* was recorded as the length from tip to base of the tallest leaf, shoot or reproductive stem of an individual.

Average maximum height for all species showed an expected initial increase in height from April through June 2002 as species developed from seedling to mature plant. Despite declining physical appearance, average maximum height of *C. javanicus*, *C. laevigatus,* and *S. virginicus* individuals remained relatively constant from July 2002 to April 2003. *B. maritimus* and *C. polystachyos* individuals, however, exhibited a decline in average maximum height from June 2002 through April 2003 (Fig. 3). This decline in average maximum height from June 2002 through April 2003 corresponds with a decline in overall physical appearance and survival of *C. polystachyos* and *B. maritimus* individuals.

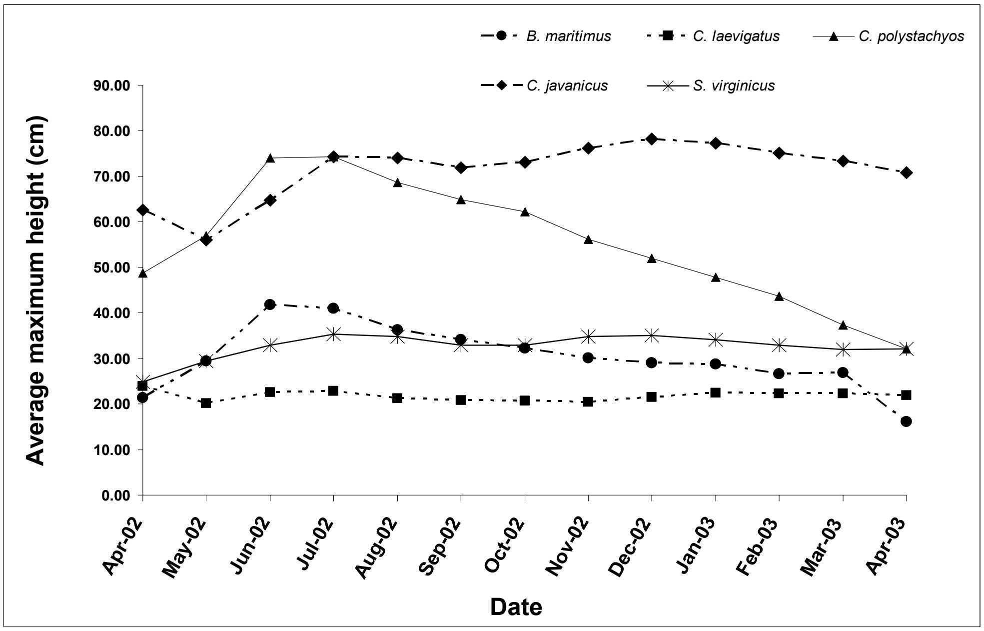


Figure 3. Average maximum height (cm) of outplanted individuals.

The number of shoots/tillers per live individual was only recorded for *B. maritimus, C. javanicus*, and *S. virginicus.* The actual number of shoots per individual was counted for *B. maritimus* and *C. javanicus.* The number of shoots per individual was estimated for *S. virginicus.* The number of shoots for both *C. javanicus* and *S. virginicus* gradually increased throughout the study period. On April 1, 2002 the average number of shoots per individual was 5.26 ± 4.12 for *C. javanicus* and less than 6 for *S. virginicus.* By the end of the study period, April 1, 2003, the average number of shoots per individual had increased to 12.58 ± 9.74 for *C. javanicus* and greater than 15 tillers per individual for *S. virginicus* (Fig. 4).

B. maritimus individuals showed an initial increase in the average number of shoots per individual from 1.06 ± 0.29 on April 1, 2002 to 3.49 ± 1.73 on June 1, 2002 (Fig. 4). However, after June the number of shoots declined to a low of 1.18 ± 0.40 shoots per live individual in mid- March 2003. However, production of new *B. maritimus* shoots began to occur in April of 2003, thus, by May 2003, the number of shoots had increased to an average of 1.38 ± 0.54 shoots per live *B. maritimus* individual. However, the average number of shoots per live individual does not indicate the incredible shoot production and then shoot die-off that occurred in *B. maritimus* planting plots. On April 1, 2002 the total number of shoots in transplanted individuals was 276. Between April and June vast production of new shoots by transplanted individuals increased the total number of shoots to 839 by June 1, 2002. New shoot production decreased rapidly at this point and many existing shoots began to senesce. By March

1, 2003 the total number of live *B. maritimus* shoots had dropped to 14. Production of new shoots from tubers of *B. maritimus* transplants increased the total number of shoots to 99 by May 1, 2003.

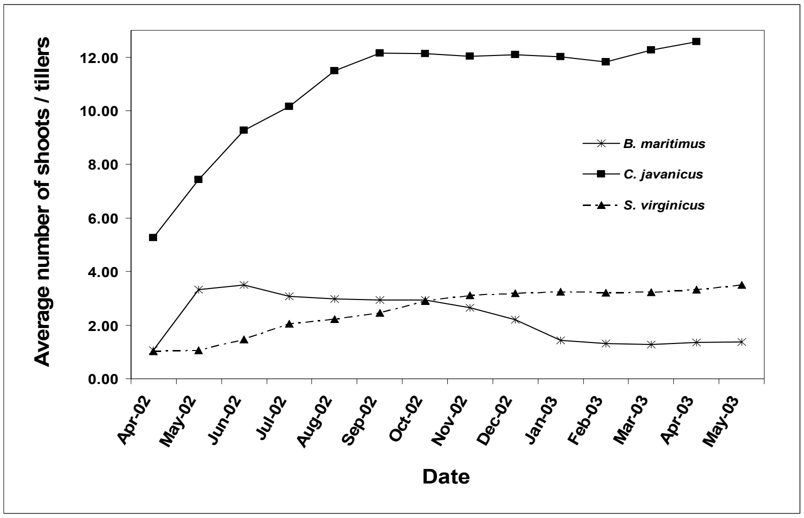


Figure 4. Average number of shoots / tillers per live individual. Numbers for *B. maritimus* and *C. javanicus* are actual counts. Numbers for *S. virginicus* are based on estimated number of tillers per individual. Estimates for *S. virginicus*: 1=1-5, 2=6-10, 3=11-15, 4=16-20, and 5=21-25 tillers per individual.

Reproduction

Reproduction in all species occurred rapidly after outplanting. By the beginning of June 2002, two months after outplanting, at least a few individuals of each species had begun flowering. By June 2002, greater than 60% of all outplanted individuals of all the sedge species, as well as *S. virginicus* had at least one reproductive shoot/tiller (Fig. 5). From May 2002 through February 2003, *C. laevigatus* and *C. polystachyos* had the highest percentage of reproductive individuals, with close to 100% of outplanted individuals bearing reproductive stems. Percent reproductive never exceeded 79% for *B. maritimus, C. javanicus,* or *S. virginicus.*

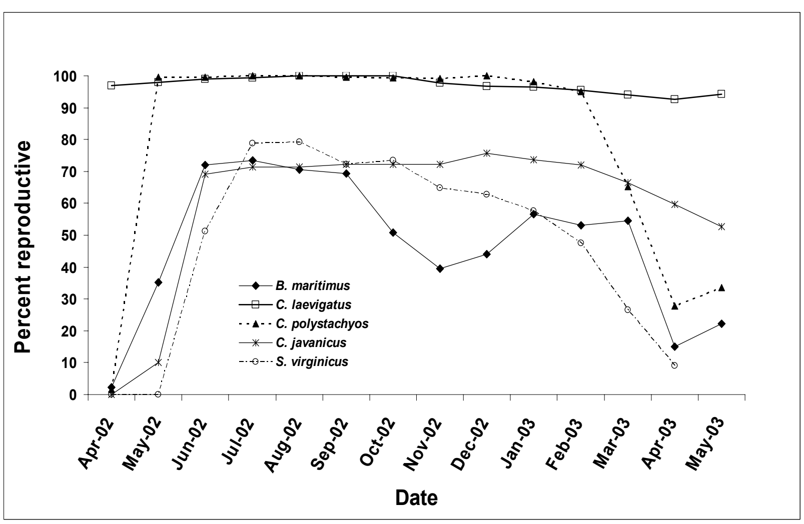


Figure 5. Percentage of outplanted individuals with at least one reproductive stem/tiller.

For all sedge species, initiation of reproduction (i.e. elongation of a reproductive stem) had begun by May 1, 2002. Three of the four sedge species (*B. maritimus, C. laevigatus,* and *C. polystachyos)* showed distinct similarities in their reproductive patterns (Fig. 6). Peak reproduction, as measured by the average number of reproductive stems per individual, for these three species occurred by July 14, 2002. After this peak, initiation of new reproductive stems declined and, thus the average number of reproductive stems per individual declined steadily from the middle of July 2002 through April 2003. Reproduction in both *S. virginicus* and *C. javanicus* exhibited a more gradual climb from June through December 2002 at which point average number of reproductive stems per individual sharply declined in *S. virginicus* (Fig. 6). From December through April 2003, the average number of reproductive stems/individual declined only slightly for *C. javanicus*. Perhaps in response to heavy rainfall in April 2003, the number of new reproductive shoots had increased for individuals of *B. maritimus, C. javanicus* and *C. polystachyos* when monitored in May 2003.

For each planting plot, final average number of reproductive stems per individual was correlated with final average health-code in *B. maritimus* ($r=0.341$; $p=0.031$), *C. laevigatus* ($r=0.824$; $p<0.0001$), *C. javanicus* ($r=0.692$; $p<0.0001$), *C. polystachyos* ($r=0.579$; $p=0.0006$), and *S. virginicus* ($r=0.450$; $p=0.0040$).

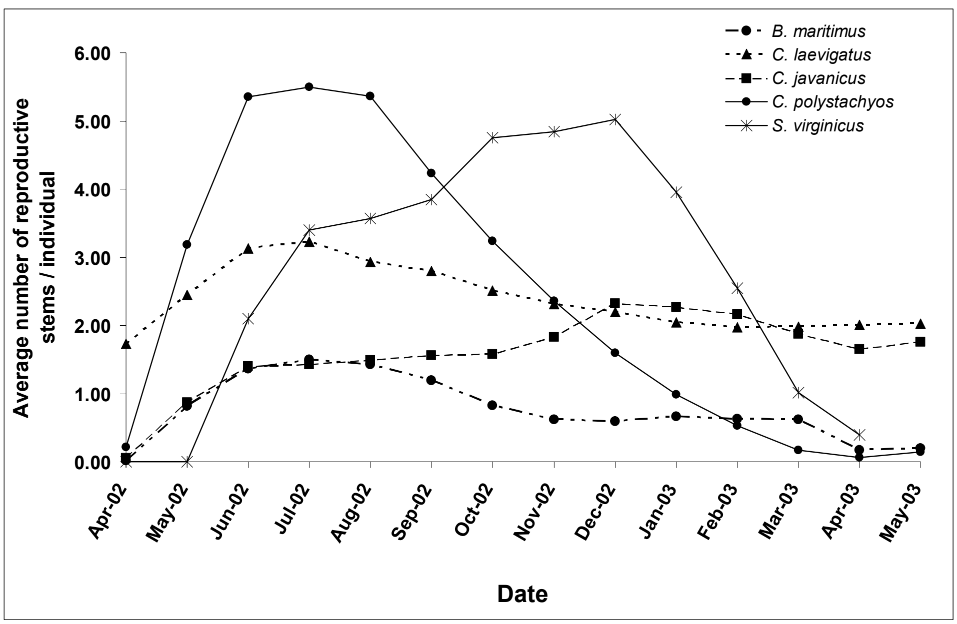


Figure 6. Average number of reproductive stems/individual. Numbers for *B. maritimus, C. javanicus, C. polystachyos,* and *S. virginicus* are based on actual counts. Numbers for *C. laevigatus* based on estimated number of reproductive stems per individual. Estimates for *C. laevigatus*: 0 = no reproductive stems, 1 = 1-10, 2 = 11-20, 3 = 21-30, 4=31-40 reproductive stems.

The remaining two species, *J. ovalifolia* and *S. portulacastrum* did not exhibit a straightforward reproductive pattern. The average number of flowers per plot for these two species fluctuated throughout the study period (Fig. 7). *S. portulacastrum* showed an initial burst of flower production in June of 2002 followed by an abrupt drop in number of flowers per plot. There was a second slight increase in flower production in October 2002 followed by a decline in the numbers of flowers per plot for much of the study. *J. ovalifolia* showed two distinct peaks of flower production, one in July 2002 and the second in February of 2003. Final number of flowers per plot was correlated with final average health-code per plot for both *J ovalifolia* and *S. portulacastrum* (r=0.440; p=0.0045 and r=0.347; p=0.031 respectively).

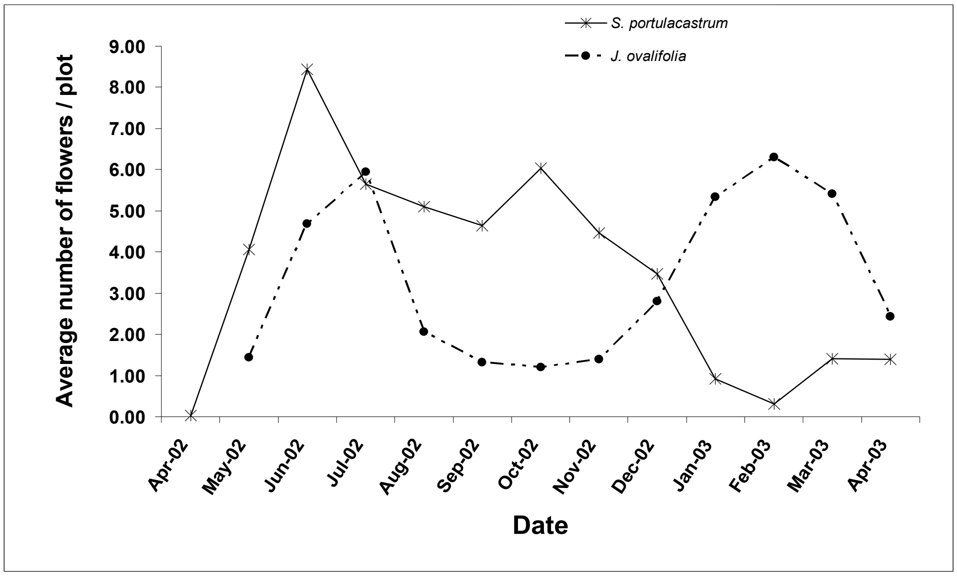


Figure 7. Average number of flowers per 0.25m^2 outplanting plot for *S. portulacastrum* and *J. ovalifolia.*

Percent Cover

Non-planted Species Composition

Percent cover of existing vegetation in outplanting plots and control plots was documented prior to planting of native species. Once percent cover was recorded, all existing vegetation was manually removed from each outplanting and control plot and planting of native species commenced. Beginning in April 2002 percent cover of all planted and non-planted species in each plot was monitored twice monthly.

Prior to outplanting, existing vegetation in native sedge outplanting and control plots (plots not planted with native sedge species) was dominated by *Pluchea indica* (Indian fleabane), *Bacopa monnieri* (`ae `ae), and *Batis maritima* (pickleweed). Almost 97% of existing vegetation in these plots was composed of only five species (Table 2). Existing vegetation in native groundcover outplanting plots was dominated by similar species, with *P. indica* and *B. maritima* providing almost 80% of total existing vegetative cover (Table 2).

Table 2. Species composition in study plots prior to outplanting (March 2002). Relative cover of dominant species in outplanting and control plots prior to outplanting of native species.

	Species						
	Pluchea indica	***Bacopa* monnieri***	***Batis maritima***	***Atriplex suberecta***	***Typha latifolia***	***Bulboschoenus* maritmus***	***Total***
Sedge plots	32.68%	30.79%	17.34%		10.23%	5.74%	96.78%
Groundcover plots	58.93%	4.99%	19.29%	9.01%	2.10%	1.74%	96.06%

* = native (indigenous) species

Shortly after outplanting, non-planted vegetation began reappearing in the study plots. Initially it was mainly native species, such as *Bacopa monnieri* (`ae `ae)*, B. maritimus* and *Heliotropium curassavicum* (kipukai) that appeared in many of the planting plots. Clearly native species initially responded well to the removal of invasive plant species such as Indian fleabane (*Pluchea indica)* and pickleweed (*Batis maritima).* However, *B. monnieri* was the only non-planted native species present in the majority of the planting plots by the end of the study.

After one year, four of the five most common species in sedge plots prior to outplanting had re-colonized and, were again among the most common species recorded in sedge planting and control plots (Fig. 8). To calculate proportion of total vegetative cover (relative cover), only percent cover from "competition" plots (plots did not have non-planted species removed once a month) were considered. The most widespread species in these competition plots at the end of the study was *B. monnieri.* Approximately 67% of total vegetative cover in all of the sedge competition plots (planting plots and control plots) was composed of *B. monnieri* (Fig.8). Re-colonization of *B. monnieri* in only control plots was even higher, with 95.1% of all vegetative cover in control plots consisting of *B. monnieri.* Interestingly, at the end of the study, *C. javanicus* had the second most abundant relative cover (11.8%) of all species (planted or non-planted) in all sedge planting and control plots. Obviously, a large portion of this cover was located in *C. javanicus* planting plots; however, relative cover of *C. javanicus* in control plots only was the third highest (7.7%) of all species recorded in these plots.

Vegetative cover of *P. indica, B. maritima,* and *T. latifolia*, after one year, had not attained pre-outplanting levels in sedge planting and control plots. Percent contributions to vegetative cover prior to outplanting were 32.7%, 17.3 % and 10.2% for *P. indica, B. maritima,* and *T. latifolia* respectively. One year post-outplanting these numbers had dropped to less than 3% for *P. indica*, and *T. latifolia* and less than 12% for *B. maritima* (Fig. 8).

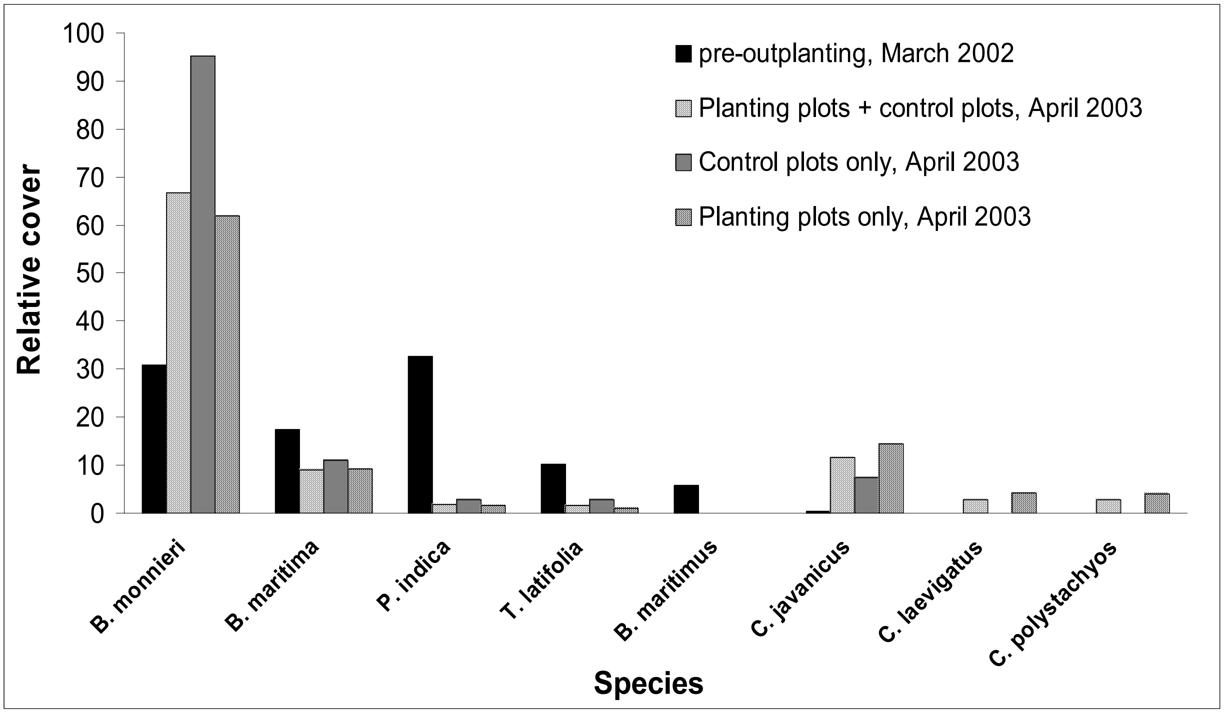


Figure 8. Overall relative cover of dominant species in sedge planting and control plots ("competition" plots only). Columns indicate relative cover prior to outplanting of native species and one year later (April 1, 2003) in: planting and control plots, control plots only, and sedge planting plots only.

A similar pattern occurred in groundcover plots (planting plots and control plots). After one year, three of the five most common species in groundcover plots prior to outplanting had re-colonized and were again the three most dominant species in groundcover outplanting and control plots (Fig. 9). However, *P. indica,* which was responsible for almost 60% of total vegetative cover in groundcover plots prior to outplanting (Table 2), did not regain this dominance after a year's time. In April 2003, *P. indica* provided 18.3% of total vegetative cover in planting + control plots and 24.1% in control plots only. As in the sedge plots, the most widespread species in groundcover plots at the end of the study was *B. monnieri.* Approximately, 32% of vegetative cover in all groundcover plots (planting plots plus control plots) consisted of *B. monnieri. B. maritima* was the third most abundant cover in all groundcover plots (planting plots plus control plots) and control plots only with 15.2 and 16.7% respectively. However, cover in groundcover planting plots (i.e. control plots not included) was dominated first by *B. monnieri* and second by *J. ovalifolia.* Plantings of *J. ovalifolia* produced dense cover in plots where seedlings had been planted. As well, *J. ovalifolia* had managed to spread into adjacent planting plots and control plots of the other groundcover species. *A. suberecta* and *T. latifolia*, which, had been the fourth and fifth most abundant species prior to outplanting, were virtually absent from planting plots and control plots in April of 2003 (Fig. 9).

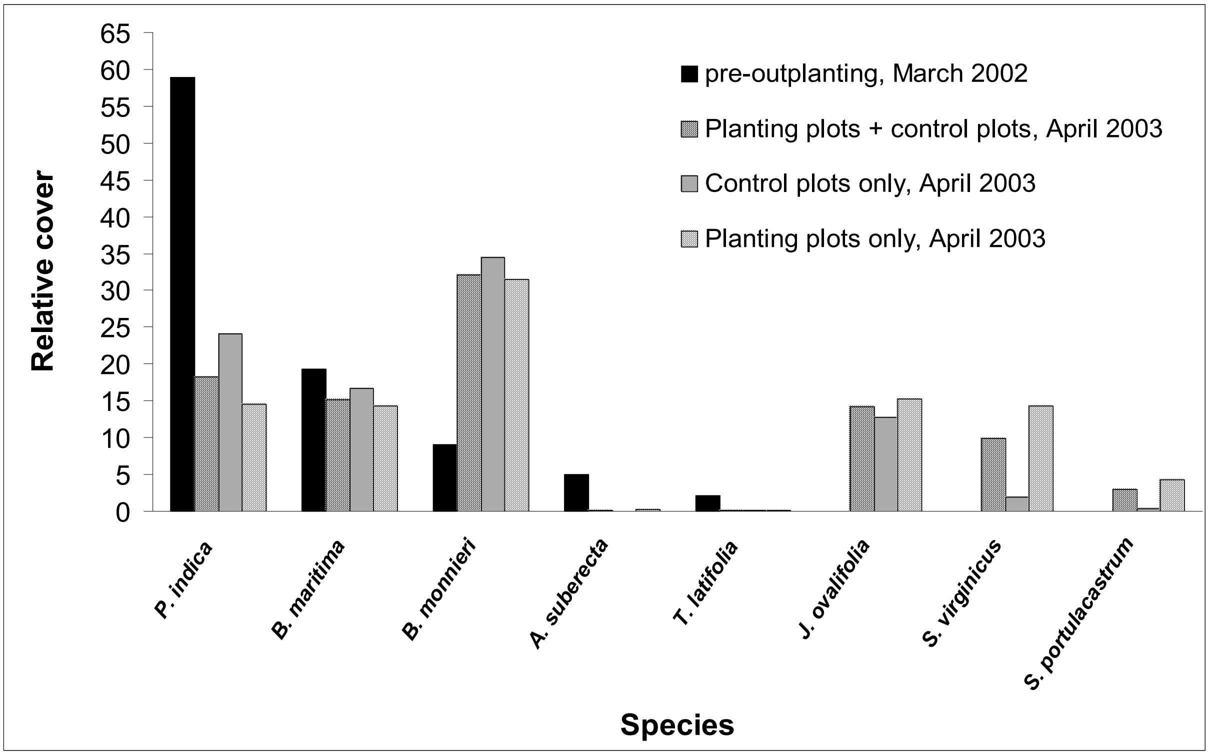


Figure 9. Overall relative cover of dominant species in groundcover planting and control plots ("competition" plots only). Columns represent relative cover prior to outplanting of native species and one year later (April 2003) in: all planting and control plots, control plots only and groundcover planting plots only.

Outplanted Native Species Composition

Shortly after outplanting, average percent cover of native species in outplanting plots ranged from 21.8% for *B. maritimus* to 60.1% for *C. polystachyos.* The range in percent cover varied primarily due to morphological characteristics of each native species. For example, *B. maritimus* has erect stems and produces relatively few linear leaves, whereas *C. polystachyos* forms many basal leaves which give the plant a somewhat bushy appearance.

The greatest average percent covers recorded during the study period were for *C. polystachyos* (85.0 ± 15.4%) in June of 2002 and *C. javanicus* (83.5 ± 17.3%) in August 2002. The lowest average percent covers during the study period were recorded for *B. maritimus* with 0.4 ± 0.9% in March 2003 and 24.5 ± 23.9% for *S. portulacastrum* in April 2003.

Table 3. Monthly average percent cover of outplanted native wetland species in planting plots. Averages are based on planting plots for each species individually, control plots not included.

Date	Species						
	Sedge Plots				Groundcover plots		
	C. polystachyos	*B. maritimus*	*C. laevigatus*	*C. javanicus*	*S. virginicus*	*S. portulacastrum*	*J. ovalifolia*
Apr-02	60.1%	21.8%	32.3%	59.8%	28.2%	42.1%	
May-02	82.4%	40.2%	40.9%	76.4%	41.5%	52.8%	25.7%
Jun-02	85.0%	43.6%	47.4%	81.4%	48.2%	56.8%	31.8%
Jul-02	81.3%	41.6%	49.7%	73.3%	53.6%	56.8%	43.9%
Aug-02	79.8%	35.6%	47.0%	83.5%	59.1%	52.6%	59.9%
Sep-02	76.6%	30.6%	46.3%	83.3%	62.3%	46.2%	69.3%
Oct-02	68.2%	25.3%	43.0%	83.4%	62.5%	43.7%	72.4%
Nov-02	56.8%	17.7%	39.1%	82.4%	64.6%	39.6%	71.6%
Dec-02	45.1%	8.3%	36.3%	82.7%	65.8%	34.8%	70.8%
Jan-03	38.2%	2.3%	4.6%	80.7%	66.9%	29.4%	64.3%
Feb-03	35.7%	1.0%	33.1%	80.2%	68.2%	27.0%	58.9%
Mar-03	33.1%	0.4%	30.7%	81.2%	69.6%	25.3%	54.1%
Apr-03	31.7%	0.7%	29.4%	81.2%	70.3%	24.5%	52.30%

Average percent cover in planting plots steadily increased for all species from April 2002 to July 2002 (Table 3; Fig. 10). From July 2002 through April 2003 average percent cover decreased for *B. maritimus, C. laevigatus, C. polystachyos,* and *S. portulacastrum*. This decline in average percent cover corresponds to declining health and survival of these four species.

Despite slightly declining health, average percent cover of *S. virginicus* continued to rise throughout the study period. Average percent cover of *C. javanicus* increased through August 2002. At this point average percent cover for *C. javanicus* plots plateaued around 83%. Average percent cover of *J. ovalifolia* steadily increased until mid-October 2002 when it reached its maximum average percent cover of 73.0 ± 15.6%. At this point average percent cover began declining in *J. ovalifolia* outplanting plots.

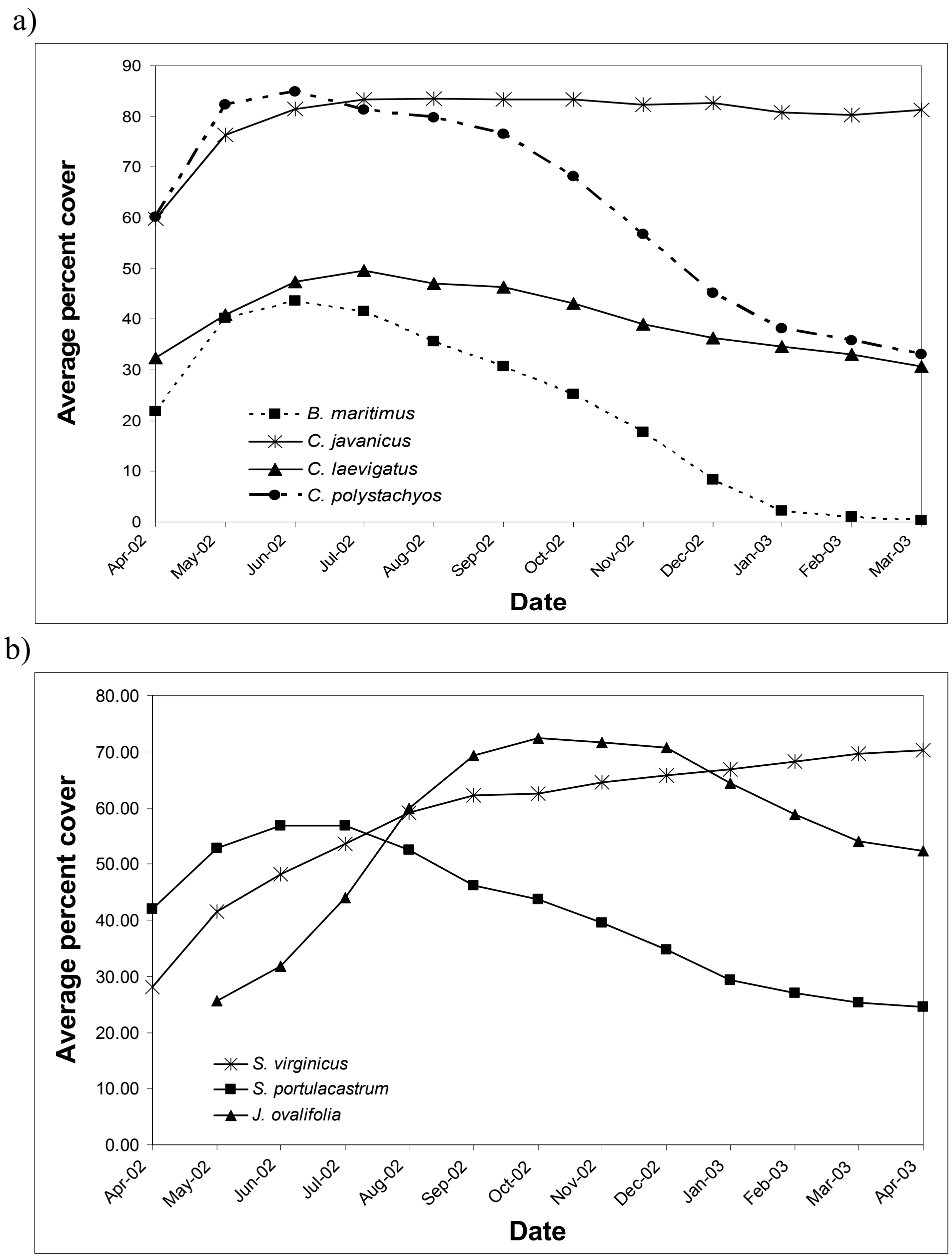


Figure 10. Monthly average percent cover in native species outplanting plots; (a) average percent cover for sedge species, (b) average percent cover for groundcover species.

III. Results and Conclusions: Individual Outplanted Native Species Accounts

Overall, the outplantings of native wetland plant species at Honouliuli were fairly successful. At the conclusion of the study, 67% of the original plants were still alive. The majority of *C. javanicus, J. ovalifolia,* and *S. virginicus* plants were healthy and robust. Outplantings of *B. maritimus, C. laevigatus, C. polystachyos,* and *S. portulacastrum* were not as successful. Although all plants in this study are at least short-lived perennials, several species, most notably *B. maritimus* and *C. polystachyos,* appeared to go through a die-back and/or senescent period beginning in late fall and continuing through winter. Beginning in late March, health and physical appearance of live individuals of many species began to improve dramatically and production of new vegetative and reproductive shoots resumed again. However, this study only followed the plants for one year, making it difficult to tell if this fall/winter decline is an annual phenological pattern or was primarily related to timing of outplanting and/or specific environmental conditions that occurred during this study period.

Species such as *C. javanicus* and *S. virginicus,* which formed tall, dense stands, were less readily overgrown by non-planted species and appear to have the highest likelihood of continued survival at Honouliuli. In sites where it survived, *C. polystachyos* also provided dense cover. Lower growing species, such as *S. portulacastrum* and *C. laevigatus,* in "competition" plots were rapidly overgrown by non-planted species such as *B. monnieri* and *B. maritima.* However, given another year's time, without any vegetation control or maintenance, all species would most likely succumb to the continued growth of species such as *B. maritima, P. indica, T. latifolia* and eventually *Leucaena leucocephala,* and *Prosopis pallida.* Periodic control of non-desired vegetation would be required to maintain populations of these native species. As all of the sedge species, as well as, *S. virginicus* can spread vegetatively through underground rhizomes and/or tubers, removal of invasive species may be all that is required to promote spread or improve the health of these species. Although no seedlings were observed in any of the planting or control plots, many of the outplanted species had high rates of seed production. It is possible that germination and seedling growth was prevented by the thick cover of *B. monnieri* in most planting and control plots. Removal of species such as *B. monnieri, B. maritima,* and *P. indica* may allow seeds of native species in the seedbank to germinate and mature, further spreading populations of native wetland species in a restoration area.

B. maritimus

a) b)

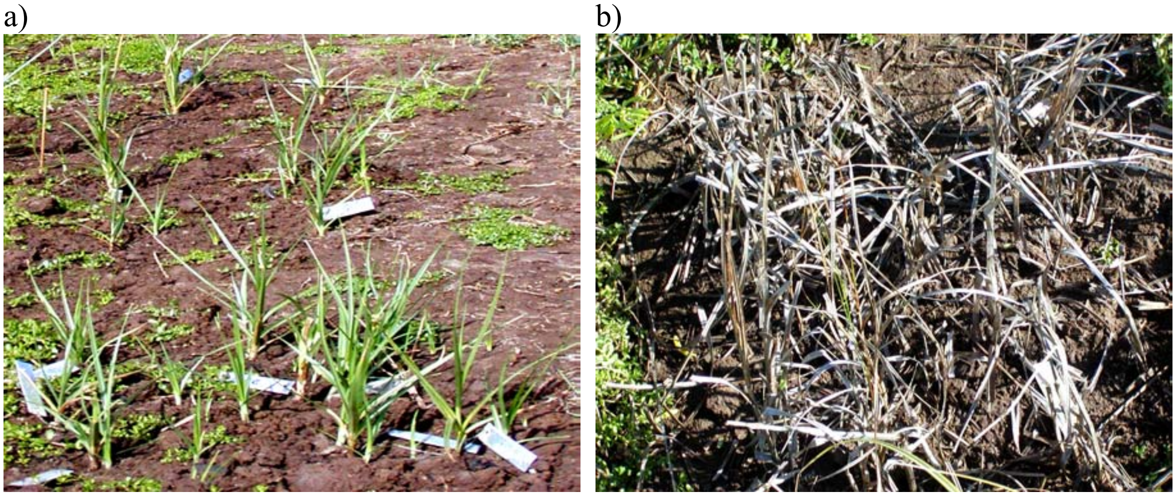


Figure 11. *B. maritimus* transplants: a) shortly after transplanting and b) in December 2002 – notice the few live stems amongst all the dead / senescent stems.

Survival and Physical Appearance

Transplants of *B. maritimus* initially flourished in their new sites at Honouliuli. However, beginning in November, *B. maritimus* shoot senescence was widespread in all planting plots (Fig. 11). By December 1, 2002 only 51.5 % of transplanted *B. maritimus* plants still had live stems. Further, this "die-back" occurred in other natural populations of *B. maritimus* at Honouliuli. By March 1, 2003 only 11 (4.2%) of the transplanted *B. maritimus* individuals had live shoots. However, beginning in late March 2003, new shoots started appearing in planting plots next to tags marking the original transplanted individuals. Presumably the underground tubers of many *B. maritimus* transplants were still alive and were beginning to produce new shoots. By May 1, 2003 seventy-two *B. maritimus* plants (27.7% of the original transplants) had produced new shoots. Annual die-back and overwintering by means of underground tubers is the typical life history pattern for *B. maritimus* in temperate climates. This may be the pattern of *B.maritimus* in Hawai`i as well. Annual winter die-back of *B. maritimus* populations, followed by production of new shoots in spring, has been noted by others working in Hawai`i wetlands (Eric Guinther, pers. comm. 2003).

The majority of newly-emerged *B. maritimus* shoots recorded in May of 2003 appeared in no-competition plots. In fact final survival rates per were significantly greater in no-competition plots than in competition plots (mean = 43 ± 23% and 15 ± 18% in no-competition and competition treatment plots respectively. Two-way *t*-test: $t=4.258$; $df=38$; $p<0.0001$). Thus, control of invasive or non-native species in areas where *B. maritimus* has been transplanted may be required for the spring re-growth of *B. maritimus.* Final percent survival was not significantly different in high-density than in low-density plots.

Physical appearance of *B. maritimus* individuals followed a similar pattern to the survival pattern described above. Initially, transplants were quite healthy and vigorous; however, by late November the average health-code had dropped to 2.30 ± 1.14 (health-code of 2 indicates >50% of individual dead or dying). Physical appearance continued to decline until mid-March 2003, at which point, production of new healthy shoots caused the health-code to climb to an average of 4.88 ± 0.34 by May 1, 2003. There were no significant differences in final average health-codes for live individuals in high-density vs. low-density plots or competition vs. no-competition plots. Final average percent survival and health-code were not correlated with soil salinity or soil temperature.

Another noticeable trend in both transplanted and natural stands of *B. maritimus* is that stands closer to open water or growing in shallow water appeared taller, thicker stemmed, darker green and generally more robust. Water level in the pond fluctuated somewhat throughout the study period, however, overall the water level continued to drop throughout the course of the year. This resulted in planting plots being much farther from the edge of the water than when outplanting occurred. At the beginning of the study, in April of 2002, water level in the pond was approximately 50 cm, but by the end of the study period, in April 2003, pond water level had dropped to 27.4 cm. *B. maritimus* planting plots that were originally 0.25 m from the water's edge were, on average, 1.57 m from the edge of the water at the end of the study. Thus, annual die-back may explain some shoot senescence seen in *B. maritimus* planting plots, but distance from open water and concurrently lowered soil moisture probably contributed significantly to poor health and permanent death of *B. maritimus* individuals.

Vegetative Growth

B. maritimus transplants had an average maximum height of 21.45 ± 6.40 cm and an average of 1.06 ± 0.29 shoots when outplanted. At the conclusion of the study the average maximum height had dropped to 16.32 ± 7.70 cm and the average number of shoots per individual was only 1.38 ± 0.54. This drop in height was primarily because the majority of live shoots at the conclusion of the study were newly emerged and emerging shoots. These shoots were not fully mature and thus had a much shorter stature than mature *B. maritimus* shoots. The peak of vegetative growth for *B. maritimus* individuals occurred in June of 2002 when the average number of shoots per individual was 3.50 ± 1.70 and the average maximum height was 41.73 ± 12.40 cm. Final average height and average number of shoots was not significantly different in low-density vs. high-density plots or in competition vs. no-competition plots.

Reproduction

Transplants of *B. maritimus* showed high reproductive rates. By July 2002, 73% of transplanted individuals had at least one shoot in some stage of reproduction (i.e. flowering, seed ripening, seed setting or seed head senescing). By May of 2003 only 22.2% of live individuals were reproductive. This is primarily due to the die-back of the majority of *B. maritimus* shoots over the winter. The majority of live shoots in May 2003 were quite young and may not have yet reached reproductive maturity. Despite low reproduction, final average number of reproductive stems was greater in no-competition plots than in competition plots (mean = 0.7±0.86 and 0.05±0.224 respectively; Mann-Whitney Rank Sum Test T=498; n=20; p=0.02). Final average number of reproductive stems was not greater in high-density than low-density plots. Similar to *C. javanicus*, despite prolific seed set, no *B. maritimus* seedlings were observed in any of the planting or control plots throughout the study period.

Percent Cover

Initial average percent cover of *B. maritimus* was 24.6 ± 3.9% in high-density plots and 13.7 ± 3.3% in low density plots. For the first three months of the study, average percent cover increased in all planting plots. Beginning in mid-June percent cover steadily decreased in all planting plots. One year post-planting average percent cover had dropped to 0.9 ± 1.2% in high-density, 0.5 ± 1.1% in low-density. By May 2003, average percent cover had risen to 3.6 ± 3.2% in high-density plots, 2.1 ± 2.1% in low-density plots. There was no significant difference between final percent cover in high-density vs. low-density plots. Final percent cover in no-competition plots was significantly greater than final percent cover in competition plots (median = 4.0 and 2.0 for no-competition and competition plots; Mann-Whitney Rank Sum Test: T= 277.5; n=20; p<0.0001). Non-planted species appear to have impeded the production of new shoots in competition planting plots (Fig. 12). Final percent cover of non-planted species was not significantly different in high-density vs. low-density vs. control plots. Stands of *B.*

maritimus did not maintain abundant cover and planting plots were readily being invaded by non-planted species.

Conclusions

a) b)

Figure 12. *B. maritimus* April 2003. a) Plot showing production of new stems after winter die-back and b) typical competition plot, *B. maritimus* transplants overgrown by *B. monnieri* and *B. maritima.*

B. maritimus is a common component in many coastal wetlands around the state, and thus is a desired species for re-vegetation projects. Natural populations of *B. maritimus* at Honouliuli form dense stands, primarily in shallow water and extending slightly in from the edge of the water. *B. maritimus* transplants at Honouliuli readily matured and reproduced from transplanted stems as long as the underground rhizome and tuber was kept intact. However, despite being culled from these healthy populations, transplanted populations of *B. maritimus* never appeared as robust or reached the density of natural populations at Honouliuli. Average percent cover was never greater than 52.3 ± 10.7% for high-density or 35.5 ± 13.7% for low-density plots. Although *B. maritimus* readily produces new vegetative shoots, it may take a while for sparse stands to fill in. Initially planting dense stands of *B. maritimus* may be preferred if dense stands are desired in a quicker time frame. Planting *B. maritimus* plants directly in shallow water may also increase shoot production, and thus increase percent cover in outplanted *B. maritimus* populations. Studies have demonstrated that the greatest numbers of shoots were produced by *B. maritimus* plants growing in shallow water (Clevering and Gulik 1997; Clevering and Hundscheid 1998). It may also be necessary to initiate vegetation control of invasive species in early spring in order to allow for spring re-emergence of *B. maritimus* shoots from underground tubers and/or allow for germination of seeds produced the previous summer and fall.

Cyperus javanicus

a)

Figure 13. *C. javanicus* a) shortly after outplanting and b) same plot 4 months later (July 2002)

<u>Survival and Physical Appearance</u>

C. javanicus plantings were one of the most successful of the outplanted species. Plants exhibited high survival and reproductive rates. In May 2003, over a year after outplanting, 96.5% outplanted *C. javanicus* plants were still alive. Further, 8 of the 9 plants that did not survive were located in one outplanting block. Five of these 8 plants were in one planting plot. High soil salinity could be a contributing factor to the high mortality rate exhibited in this area. Soil salinity in this block, with an overall yearly average of 11.87 mmhos (9.49 ppt), was the highest of the *C. javanicus* planting blocks. Final percent survival was negatively correlated with average soil salinity ($r = -0.743$, $p = 0.014$). However, *C. javanicus* individuals did survive in a wide range of average soil salinities ranging from a minimum of 3.91 mmhos (2.50 ppt) to a maximum of 11.58 mmhos (9.26 ppt). It appears as though there may be a threshold of soil salinity, above which, *C. javanicus* individuals may not be able to survive.

After one year's time the majority of *C. javanicus* individuals appeared quite healthy and robust; 85.5% of the surviving plants on May 1, 2003 had a health-code of 3 or above. However, there were several planting plots in which the majority of *C. javanicus* individuals appeared stunted, chlorotic, and generally unhealthy. Average health-code for entire planting plots ranged from 1.25 to 5. Although final average plot health-code was negatively correlated with average soil salinity (-0.343, $p=0.03$), plants in some areas with low average soil salinity measurements appeared quite unhealthy. Further, *C. javanicus* individuals were able to thrive in some areas with higher soil salinities. Presumably, soil salinity plays a role in limiting physical health of *C. javanicus* individuals, but there are clearly other environmental parameters which influence the physical health of *C. javanicus* populations.

Final percent survival and average health-code was not significantly different in high-density vs. low-density treatment plots or in no-competition vs. competition treatment plots.

Vegetative Growth

Individuals of *C. javanicus* rapidly began producing new vegetative shoots after outplanting. The initial number of shoots averaged 5.26 ± 4.12 per individual. Within three months, this number had almost doubled to 10.16 ± 5.52 shoots per individual. Production of new shoots continued throughout the study, although production slowed between September 2002 and March 2003. One year after outplanting the average number of shoots per individual had risen to 12.58 ± 9.74. However, this average does not reflect the incredible range of shoot production by *C. javanicus* individuals. Individuals exhibiting poor health had as low as one shoot, whereas the healthiest individuals showed incredible growth and had up to 88 shoots. Average maximum height for individuals over the length of the study was 68.70 ± 3.53 cm. Initial average maximum height was 62.56 ± 12.95 cm and final average of maximum height was 70.76 ± 36.31cm. Final average number of shoots and average maximum height was not significantly different in low-density vs. high-density plots or competition vs. no-competition plots.

Reproduction

By June 1, 2002, two months after outplanting, almost 70% of *C. javanicus* plants had produced at least one reproductive stem. Although percent reproductive individuals averaged between 70 and 76% from mid-June 2002 through February 2003, production of new reproductive stems slowed sharply between July and November 2002. The majority of reproductive stems during this period were in the seed ripening, seed setting and senescent stages of reproduction. There was a slight surge in production of new reproductive stems in November and December of 2002, but, once again, from January through April 2003, there were essentially no new reproductive stems being produced. During the last monitoring period in May 2003, another large increase in the number of new reproductive stems was documented. At the conclusion of the study the average number reproductive stems was 1.65 ± 2.68. Final average number of reproductive stems was not significantly different in low-density vs. high-density plots or in competition vs. no-competition plots.

Despite the large reproductive output, very few seedlings of *C. javanicus* were seen in planting plots or control plots throughout the study period. Spread of *C. javanicus* was primarily through vegetative growth via production of new shoots from underground rhizomes. Although vegetative reproduction increases plant cover in the immediate area, expansion from the immediate area is slow and limited. Given the right environmental conditions, *C. javanicus* seeds produced by outplanted plants may germinate and increase the distribution of *C. javanicus* populations. Germination of these seeds, however, may be hindered by existing dense vegetative cover by species such as *B. monnieri, P. indica,* and *B. maritima*.

Percent Cover

Cover of *C. javanicus* increased considerably in both low density and high density plots over the course of the study period. Initial percent cover was, on average, 38.7 ± 7.7 in low-density plots and 62.4 ± 10.2 in high-density planting plots. One year later, average percent cover was 73 ± 26.9 in low-density plots and 89.4 ± 21.3 in high-density plots. Excluding plots in Block 4, which experienced high mortality of *C. javanicus* individuals, low-density plots averaged 76.5 ± 26.1 percent cover while high density plots averaged 95.8 ± 7.6 percent cover.

There was no significant difference between final percent cover in low-density vs. high-density plots. Both low-density and high-density planting plots of *C. javanicus* were able to spread and provide dense cover in the planting plots. There was also no significant difference between final percent cover in competition plots vs. no-competition plots. Colonization of non-planted species did not appear to affect the final percent cover of *C. javanicus* to any measurable extent. But, there was also no difference in the final percent cover of non-planted species in control, low or high-density competition plots. Higher densities of *C. javanicus* did not appear to preclude establishment of non-planted species to a greater extent than did control or low-density plots. However, this was primarily due to the ability of *B. monnieri* to colonize low and high-density competition plots of virtually all species.

Conclusions

a)

b)

Figure 14. *C. javanicus*, November 2002. a) High-density / competition plot and b) low-density / competition plot.

C. javanicus managed to survive and thrive in a variety of conditions at Honouliuli. Further, many outplanted individuals rapidly reproduced and spread vegetatively resulting in dense cover in outplanting plots. Even lower density plantings quickly spread and provided abundant cover in outplanting plots. Final percent survival and average physical health, number of shoots, maximum height, and number of reproductive shoots for *C. javanicus* individuals in low vs. high-density and competition vs. no-competition treatments were not significantly different. Thus, even when subjected to competition from invasive species and planted in lower densities, *C. javanicus* may be able to survive, grow and reproduce. However, periodic vegetation control, especially of woody species such as *P. indica,* would be necessary to ensure the continued success of *C. javanicus* populations. This species could be a valuable component to future restoration efforts in both brackish and freshwater wetlands in the state.

Cyperus laevigatus

Figure 15 panel labels: a) (left photo) and b) (right photo).

Figure 15. *C. laevigatus* planting plots. a) Shortly after outplanting in April 2002 and b) same plot one year later in April 2003.

Survival and Physical Appearance

Prior to outplanting many *C. laevigatus* plants delivered from the nursery appeared weak and spindly. Within one month after outplanting, despite a slight drop in average maximum height, outplanted individuals, appeared much more stout and healthy. Percent survival remained greater than 90% until late-October. After one year, however, outplantings of *C. laevigatus* have had only limited success. Although 76% of the original outplanted individuals were still alive one year after outplanting, the physical appearance of the majority of the plants was quite poor. In fact 61% of live plants had a health-code of 2 or less on April 1, 2003. The poorest overall health for *C. laevigatus* individuals occurred in early March of 2003 when the average health-code dropped to 2.19 ± 1.02. Physical appearance did begin to improve in mid-April, perhaps due to the onset of heavy rains in March. By May 2003 the average health-code had climbed slightly to 2.76 ± 1.21.

There was quite a range in physical appearance and percent survival of *C. laevigatus* which tended to correspond with planting area. Average health-codes for entire planting plots ranged from 1.8 to 4.3. So, in some areas *C. laevigatus* plants were healthy and robust, where in other areas around Pond 2 plants were small and chlorotic. Further, mortality in just three of the ten planting blocks accounted for 58% of total mortality in *C. laevigatus.* High soil salinity may have contributed to the mortality and poor health observed in *C. laevigatus.* Planting plots in Block 4, the planting block with the highest death rates and lowest final health-code, also had the highest average soil salinity (12.34 ± 0.56 mmhos). However, planting plots in a nearby block, which had the second highest soil salinity (overall average 12.07 ± 1.24 mmhos), had a 93% survival rate. Individuals in this block also appeared quite healthy and robust (average health code = 3.85). Overall average soil salinity was significantly negatively correlated with final percent survival in each planting plot ($r = -0.379$, $p=0.016$), but was not correlated with final average health in each planting plot. There was no significant difference in final average percent survival or average health-code in low-density vs. high-density or competition vs. no-competition treatment plots.

Vegetative Growth

Individuals of *C. laevigatus* produced too many shoots too accurately and efficiently count, so this parameter was not measured for these individuals. Average height of *C. laevigatus* individuals did not fluctuate much during the study period. The lowest average maximum height of 19.56 ± 5.24 cm was recorded on April 14, 2002. The greatest average maximum height, 23.93 ± 6.46 cm, actually occurred at the time of outplanting. For the duration of the study average maximum height fluctuated around 21 cm. This is much shorter than populations occurring elsewhere in the state. According to Stemmermann (1981), height of *C. laevigatus* individuals ranges from 10 to 70 cm. In April 2003, at the conclusion of the study, only 26% of live plants (60 of 229) had a maximum height of 25 cm or greater. The final average maximum height was 21.92 ± 6.73 cm. Final average height was similar in low-density and high-density treatment plots and competition and no-competition treatment plots.

Reproduction

Outplanting stock of *C. laevigatus* was propagated from divisions of mature nursery plants. Thus, many plants already had reproductive stems prior to outplanting. In fact at the beginning of the study 97% of *C. laevigatus* individuals had at least one reproductive stem. This prolific reproduction continued throughout the study period. Percent reproductive individuals never dropped below 92% and from August through November 2002 100% of *C. laevigatus* plants had at least one reproductive stem. Number of reproductive stems was estimated for *C. laevigatus.* Average number of reproductive stems per plant ranged from greater than 10 in April 2002 to greater than 30 in July of 2002. At the end of the study the average number of reproductive stems was just greater than 20 per plant. As in all other species no seedlings were observed in planting plots or control plots during the study period. Spread of *C. laevigatus* appeared to be primarily through vegetative growth. Final average number of reproductive stems was not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

Percent Cover

Initial average percent cover of *C. laevigatus* was 35.6 ± 8.6 in high-density plots and 19.6 ± 6.8 in low-density plots. Cover of *C. laevigatus* initially increased for both high-density and low-density plots. Maximum average cover for high-density plots (60.3 ± 12.2%) and low-density plots (39.4 ± 14.6%) occurred in July of 2002. Beginning in early August, due to increased mortality, decreased health, and increased competition by non-planted species, average percent cover of *C. laevigatus* began to decline. One year after outplanting, average percent cover had decreased to 34.3 ± 24.7 for high-density plots and 25.4 ± 17.9 for low-density plots. This difference in final average percent cover between high-density and low-density plots was not statistically significant. Final average percent cover in no-competition plots was 33.9 ± 21.3 and

24.8 ± 21.6 in competition plots. This difference was also not significant. Final percent cover of non-planted species was similar in control, low-density and high-density competition plots. Similar to *C. javanicus,* this was primarily due to the ability of *B. monnieri* to readily colonize and form dense cover in planting plots, regardless of the density of *C. laevigatus.*

Conclusions

a)

b)

Figure 16. *C. laevigatus* planting plots in April 2003. a) Relatively healthy plants in planting plot with high soil salinity measurements and b) lack of abundant cover was noticeable in many planting plots.

Outplantings of *C. laevigatus* were only moderately successful. Interestingly, the planting block with the healthiest, most robust plants (based on average of health-code for all plants in all planting plots within a block) had the lowest average soil salinity, but the blocks with the second through fourth highest final average health-codes were the planting blocks with three of the four highest average soil salinity measurements. Based on this evidence, although soil salinity was slightly negatively correlated with survival, it does not appear to be the primary cause of mortality in *C. laevigatus.* Clearly, *C. laevigatus* is able to tolerate and thrive under moderate levels of soil salinity. Some of the healthiest plants were found growing in plots with an average salinity of 12.06 mmhos (9.65 ppt). Further, robust stands of *C. laevigatus* are found growing in other brackish and saline wetlands around the state. *C. laevigatus* may be a potential candidate for re-vegetation in areas under the influence of moderately high salinity.

As soil temperature and final distance from water were not correlated with final *C. laevigatus* health and survival, and plants were shown to tolerate salinity, there are clearly other environmental factors not examined by this study that have inhibited *C. laevigatus'* ability to thrive at Honouliuli. Other factors such as soil moisture, nutrient levels, and/or soil type may have played a role in limiting the success of *C. laevigatus* outplantings. In a study investigating the potential use of *C. laevigatus* for wastewater treatment, Van Dyke (2001) observed that *C. laevigatus* plants subjected to higher nutrient levels were substantially taller and darker green than those subjected to lower nutrient levels.

During this study very few *C. laevigatus* planting plots achieved and maintained abundant cover. Planting *C. laevigatus* in higher densities initially may increase the likelihood of establishing persistent dense stands of *C. laevigatus.* Until dense stands are attained, *C. laevigatus* outplanting areas may require removal of invasive plants in order for the plants to thrive. Planting *C. laevigatus* at the water's edge and/or in shallow water could also potentially increase survival rates and improve health of this species.

Cyperus polystachyos

a) **b)**

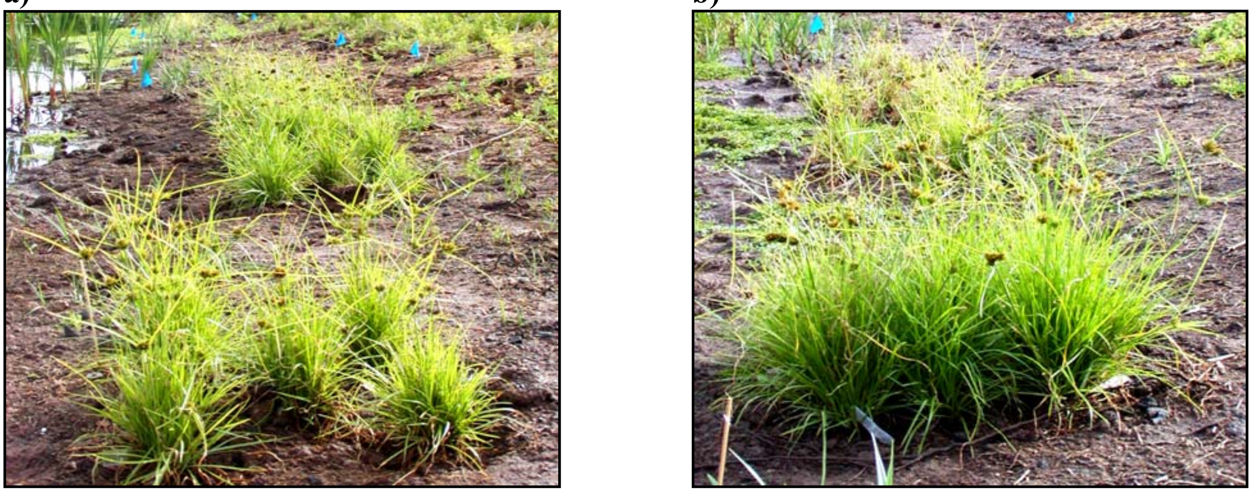


Figure 17. *C. polystachyos* shortly after outplanting. a) Low-density and b) high-density plot.

Survival and Physical Appearance

Initially, the outplanting of *C. polystachyos* appeared as though it would be quite successful. For the first six months of the study, plants were very healthy, reproduced prolifically and grew in both stature and diameter. In fact in the beginning of September 2002 more than 96% of *C. polystachyos* plants were still alive. Although physical appearance of *C. javanicus* started rapidly declining by August 2002, it was not until mid-November that survival rates sharply declined. This decline in survival continued through the end of the study. One year after outplanting only 173 of the original 300 plants (57.7%) were alive. Almost 50% of final mortality occurred in 2 planting blocks and more than 60% of final mortality occurred in 3 planting blocks. All individuals in planting plots in blocks 3 and 4 died before the end of the study. Block 4, on the east side of Pond 2, (See Appendix 1: Map of Outplanting Field Trials) was the site of high mortality of both *C. javanicus* and *C. laevigatus* as well. Similar to *C. javanicus* and *C. laevigatus,* average soil salinity recorded in block 4 (12.16 mmhos / 9.73 ppt) was the highest of the *C. polystachyos* outplanting blocks. Average soil salinity in block's 3 and 5, the other blocks with high, were 8.08 mmhos (6.46 ppt) and 11.25 (9.0 ppt) respectively. Final average survival rates for *C. polystachyos* occurred in planting plots with lower average salinity measurements. Not surprisingly, final percent survival in *C. polystachyos* planting plots was strongly negatively correlated with average soil salinity ($r = -0.723$, $p<0.0001$).

Physical appearance of *C. polystachyos* individuals started to decline rapidly by the beginning of August 2002. This decline continued through March 2003. Production of new vegetative and reproductive shoots resumed in mid-April, after a period of virtually no new production, causing an increase in the average health-code at this point. In fact average health-code of *C. polystachyos* individuals increased from a low of 1.46 ± 0.79 in mid-February 2003 to 2.36 ± 1.35 by May 2003. There were distinct outplanting blocks/areas where *C. polystachyos* plants were much darker green, taller and generally healthier in appearance. As noted above, these tended to be areas with lower soil salinity. Final average health-code in each planting plot was also negatively correlated with soil salinity ($r = -0.675$, $p<0.0001$).

Final average percent survival and average health-code was not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

Vegetative Growth

Outplanted individuals of *C. polystachyos* produced a large number of shoots, so the number of shoots per individual was not recorded for this species. Maximum height and percent cover were used to measure vegetative growth and spread of *C. polystachyos.*

Height of *C. polystachyos* plants increased dramatically in the first two months after outplanting. At the time of outplanting, the average maximum height of C. *polystachyos* seedlings was 48.79 ± 5.15 cm. By June 1, 2002 the average maximum height had increased to 73.99 ± 17.49 cm. In fact some plants were so tall their stems started to fall over. However, average maximum height started declining by mid-July as the health of *C. polystachyos* plants started to decline. By the end of the study average maximum height of live plants had dropped to 32.14 ± 12.12 cm. Presumably, this average would have, once again, started to rise with the increase in production of new shoots and reproductive stems that began in April 2003. Final average maximum height was not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

Reproduction

Reproductive stems began to develop in the majority of *C. polystachyos* plants shortly after outplanting. By May 1, 2002, 99% of outplanted individuals had produced at least one reproductive stem. By early June 100% of plants were reproductive. Although greater than 99% of individuals possessed reproductive stems through mid-December, these stems contained primarily ripening and senescing seed heads. Production of new reproductive stems basically halted in early August. As reproductive stems matured and senesced, the average number of reproductive stems per live individual declined from a high of 56.36 ± 28.20 to a low of 0.70 ± 1.19 in late March 2003. Production of new reproductive stems began to recur in mid-April 2003. This rise in reproduction at the end of the study increased the average number of reproductive stems/individual to 1.71 ± 3.0 by mid-May. As with other outplanted species, no seedlings were observed in control or planting plots during the study period. Populations of *C. polystachyos* rarely spread much beyond the edges of the original outplanting plots.

Final average number of reproductive stems was not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

Percent Cover

Healthy *C. polystachyos* plants provided dense cover in both high-density and low-density plots. Initial average percent cover was 70.4 ± 11.5 for high-density plots and 42.3 ± 8.0 for low-density plots. By mid-May average percent cover had risen to 92.1 ± 14.0 for high-density and 77.2 ± 14.6 for low-density plots. By June it was apparent that low-density plots (5 plants per 0.25 m^2) provided dense cover and high-density plots (10 plants per 0.25 m^2) were too dense. In fact, between June and October individuals in most high-density plots were so crowded that the shoots, which had grown quite tall, presumably due to overcrowding, were so tall that they were falling over. Plants in the less crowded, low-density plots were shorter, stouter and more robust.

Average percent cover for all plots rose until mid-June at which point percent cover began to decline. However, average percent cover for high-density plots did not drop below 80% and low-density plots did not drop below 70% until mid-September. This decline in percent cover appears to be more a function of declining health rather than increased mortality, as percent survival did not drop below 90% until November of 2002. Final average percent cover was 32.1 ± 34.8 for high-density and 31.2 ± 33.0 for low-density plots. Final percent cover, excluding planting plots with 0% survival, was, on average, 42.8 ± 34.0 for high-density plots and 41.5 ± 32.0 for low-density plots. Low-density plots with no-competition had the highest final average percent cover (54.0 ± 37.0) when excluding plots with 100% mortality.

There was no significant difference in final average percent cover in density or competition treatments. There was also no significant differences in final percent cover of non-planted species in high-density, low-density or control competition plots.

Conclusions

a)

b)

Figure 18. *C. polystachyos* planting plots April 2003. a) Low-density plot showing new growth that began in early April and b) complete death in low-density planting plot.

Success of *C. polystachyos* outplanting was tied to location at Honouliuli. The healthiest plots were located on the southwest corner of Pond 2. This area is closest to the freshwater input to the pond. Not surprisingly, the three areas (blocks) with the highest final survival rates and final average health-codes had the lowest average soil salinities. Close to 100% mortality was exhibited in planting plots on the east side of the pond, where higher average soil salinities were recorded. The plots with healthy individuals provided incredibly dense cover and exhibited high reproductive output. Provided that the area is favorable to *C. polystachyos,* even lower density plantings should rapidly fill in and provide abundant cover of this species. *C. polystachyos* would be a valuable component to freshwater or slightly brackish restoration efforts. This species, due to its height and bushy nature, can tolerate some competition from other species. Given the right environmental conditions, *C. polystachyos* may require less maintenance, in the form of non-native vegetation control, than other species in order to survive and thrive.

Jacquemontia ovalifolia

a) b)

Figure 19. *J. ovalifolia* outplanting block. a) After outplanting in May 2002 and b) same planting block in August 2002. Block almost entirely covered by *J. ovalifolia.*

Survival and Physical Appearance

Outplanting of *J. ovalifolia* occurred in May 2002. Greater than 90% of outplanted individuals survived through September 2002. Beginning in October 2002, survival gradually declined for the duration of the study. Approximately 70% of the original 300 plants were still alive on April 1, 2003. Unlike in many other species, mortality was randomly distributed among planting blocks and planting plots. Final overall percent survival was higher in no-competition plots (75.5%) than in competition plots (69%) and higher in low-density plots (75%) than in high-density plots (69.5%). However, the differences in final percent survival were not significantly different between competition and no-competition treatment plots or between high density and low-density treatment plots. Soil salinity and soil temperature were not correlated with final survival or average health-code of *J. ovalifolia.* Plants were equally able to survive in planting areas with high and low soil salinity and soil temperature.

Physical appearance of *J. ovalifolia* dropped very slightly after outplanting, but then remained steady from June through August 2002. Average planting plot health-code in August was 4.67 ± 0.58. Physical appearance dropped sharply from August to November 2002 and then declined more gradually from November 2002 through April 2003. Despite overall declining health, the majority of *J. ovalifolia* plants were relatively healthy at the end of the study period. More than 63% of planting plots had an average health-code of 3 or greater on April 1, 2003. Planting plot health-code at this point averaged 2.80 ± 0.99.

Final percent survival and average health-code were not significantly different in low-density versus high-density treatment plots or competition versus no-competition treatment plots.

<u>Vegetative Growth</u>

Due to its pattern of growth, (i.e. prostrate, roots at the nodes) maximum height and number of shoots was not recorded for *J. ovalifolia.* Vegetative growth of *J. ovalifolia* was measured solely through percent cover.

<u>Reproduction</u>

Reproduction of *J. ovalifolia* was measured by counting the number of flowers per plot versus per individual. Two distinct flowering peaks were exhibited in *J. ovalifolia* planting plots during this study period. The first peak in reproductive output occurred shortly after outplanting. Average number of flowers per plot rose from an average of 1.45 ± 1.34 at the time of outplanting to an average of 7.73 ± 5.69 on July 1, 2002. Flowering production slowed after July and reproductive output dropped to an average of 1.15 ± 2.89 flowers / plot in October 2002. Very few flowers were produced in October or November 2002, but flower production surged again from December 2002 through February 2003. Average number of flowers per plot in mid-February 2003 was 6.35 ± 5.68. At this point reproduction slowed again and number of flowers declined steadily from the end of February 2003 to April 2003. It would be interesting to monitor outplanted populations for another year to determine if a cyclical pattern of flower production in *J. ovalifolia* would continue to occur.

Final average number of reproductive stems was not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

<u>Percent Cover</u>

Initial cover of *J. ovalifolia* in outplanting plots averaged 32.4 ± 5.3% in high-density plots and 19.0 ± 3.2% in low-density plots. *J. ovalifolia* plants rapidly spread throughout entire planting blocks. In fact by mid-August growth of *J. ovalifolia* from planting plots had extended into 95% of the control plots. Average percent cover rose steadily from the end of May 2002 through October 2002. By November 2002 average percent cover had risen to 44.3 ± 30.3 in control plots, 65.6 ± 16.73 in low-density and 77.8 ± 20.4 in high-density plots. Final average percent cover of *J. ovalifolia* was similar in all density treatments, 51.8 ± 27.0 in control, 49 ± 18.7 in low-density, and 55.6 ± 19.5 in high-density plots. Declining health and a slight general die-back in many plots resulted in this drop in final percent cover. However, there was still abundant cover of *J. ovalifolia* in the majority of planting blocks.

There was no significant difference in final average percent cover in high-density, low-density or control plots. There was also no significant difference between final average percent cover in no-competition and competition treatment plots. Although average percent cover of non-planted species was greater in control plots and low-density plots than in high-density plots this difference was not statistically significant.

<u>Conclusions</u>

a)

b)

Figure 20. *J. ovalifolia* high-density / no-competition plots in April 2003. a) High-density plot suffering die-back in outplanted individuals and b) healthy individuals suffering no die-back.

After one year, *J. ovalifolia* outplantings appeared quite successful. Not only had 70.7% of outplanted individuals survived, but also the majority of these surviving plants looked quite healthy. *J. ovalifolia* has become established in many of the areas in which it was planted and has spread into adjacent control plots and planting blocks. *J. ovalifolia* plants tolerated, and even thrived, in a range of environmental conditions that it was exposed to in planting plots at Honouliuli.

Although this species provided relatively dense cover whether initially planted in low or high-densities, without periodic vegetation control, especially of woody-species such as *B. maritima, P. indica, P. pallida,* and *L. leucocephala,* this species would eventually be overgrown and would possibly die out. *J. ovalifolia* plants, however, prolifically set seed. These seeds may be maintained in the seedbank and removal of invasive species, such as *P. indica* and *B. maritima*, might allow these seeds to germinate. This would help maintain current populations of *J. ovalifolia* and allow for further spread of this species at Honouliuli. Dense growth and encroachment of *B. monnieri* did not seem to affect survival and growth of *J. ovalifolia* compared to its effect on other species. *J. ovalifolia* plants managed to grow on top of the thick mats of *B. monnieri*, as opposed to species such as *S. portulacastrum,* which ended up being smothered by *B. monnieri.* Due to its ease of propagation and ability to survive with fairly minimum maintenance, this species may be useful to wetland restoration projects elsewhere in the state.

Sesuvium portulacastrum

a) b)

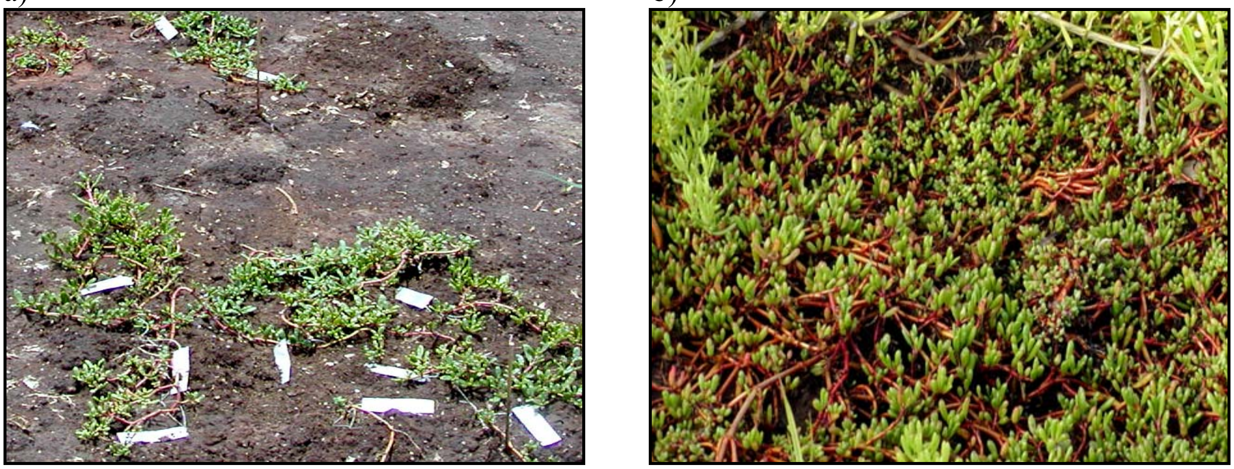


Figure 21. *S. portulacastrum* planting plot a) April 2003 and b) same plot in April 2003

Survival and Physical Appearance

Survival of *S. portulacastrum* was greater than 90% for the first six months after outplanting. Beginning in October 2002, however, survival steadily decreased for the next six months. One year after outplanting overall survival had dropped to 65% and appeared as though it may continue to drop. As opposed to *J. ovalifolia,* mortality in *S. portulacastrum* appeared tied to location. Forty-one percent of final *S. portulacastrum* mortality was located in two planting blocks and 70% of mortality was located in four of the ten planting blocks. However, unlike in many of the other species, the planting areas with high *S. portulacastrum* mortality were not the areas with the highest soil salinity or highest soil temperatures. In fact the planting areas with the highest final percent survival were the planting blocks with the highest overall average salinities. *S. portulacastrum* is considered a halophyte and is often found growing near the coast in areas exposed to saltwater or salt spray (Stemmermann 1981). It is unclear why there was such high mortality in *S. portulacastrum* and why mortality was restricted to these particular planting blocks versus being randomly distributed among all planting blocks.

Despite high survival rates for the first six months, physical appearance of *S. portulacastrum* began to decline rapidly after outplanting. By the end of June, although survival was at 98%, the majority of *S. portulacastrum* plants were in poor health. Average plot health-code dropped to its lowest level, 1.62 ± 0.80, by the end of January 2003. Beginning in February 2003 physical appearance of surviving plants improved slightly. By the end of the study average health-code per plot had risen to 2.00 ± 1.18. This rise in average health-code could partially be due to the death of plants with low health-codes in previous months and/or possibly a response to increased rainfall in January. However, by the end of the study many live plants did not appear as though they would continue to survive much longer.

Final average percent survival and average health-code were not significantly different in low-density and high-density treatment plots or competition and no-competition treatment plots.

Vegetative Growth

Due to its pattern of growth, (i.e. prostrate, roots at the nodes) vegetative growth of *S. portulacastrum* was measured solely through percent cover.

Reproduction

Reproductive output of *S. portulacastrum* was measured by counting the number of flowers per plot. One month after outplanting there was a total of only 21 flowers in all outplanting plots, for an average of 0.5 ± 1.2 flowers per plot. By the end of June flower production had increased dramatically to an average of 9.8 ± 8.8 flowers per plot. Reproduction declined from the end of June through September of 2002. A surge of reproduction occurred in October, when average number of flowers per plot rose from 4.1 ± 7.1 on September 1, 2002 to 6.5 ± 11.5 on October 1, 2002. Reproduction sharply declined from October 2002 to its lowest point, 0.2 ± 0.8 flowers/plot, in February 2003. Reproduction increased again from February through April 2003. At the conclusion of the study, April of 2003, average number of flowers was 1.4 ± 3.1 per plot. Final average number of flowers per plot was not significantly different in low-density versus high-density treatment plots or competition versus no-competition treatment plots.

This pattern of reproduction, except for the slight surge of flower production in October, was similar to the pattern observed in *C. laevigatus, B. maritimus,* and *C. polystachyos.* Each of these species exhibited an initial surge in reproduction in June or July of 2002, followed by a decline in reproduction from July2002 through February / March of 2003. At this point, reproduction in all of these species rose again from February/March 2003 through the end of the study period. As with the other species it is difficult to tell if this reproductive pattern witnessed in *S. portulacastrum* is a seasonal pattern or is related to other factors.

Percent Cover

Abundant cover of *S. portulacastrum* was achieved in relatively few outplanting plots. Initial cover of *S. portulacastrum* in outplanting plots averaged 52.3 ± 10.9% in high-density and 31.8 ± 8.4% in low-density plots. Percent cover increased steadily from April to mid-July 2002. At this point average percent cover of *S. portulacastrum* was 69.2 ± 16.8 in high-density and 45.1 ± 16.7 in low-density plots and 0.7 ± 2.0 in control plots. However, from August 2002 through the end of the study average percent cover gradually declined. Poor health and low survival in *S. portulacastrum* resulted in low cover in many outplanting plots. Final average percent cover was 34.5 ± 28.6 in high-density, 14.5 ± 12.4 in low-density, and 2.8 ± 6.7 in control plots. Final average percent cover was significantly greater in high-density plots than in low-density plots ($t = 2.87$; df = 26; $p = 0.008$).

Final average cover in no-competition plots was 30.7 ± 24.9%, and 18.3 ± 22.0 in competition plots. Final average percent cover in no-competition planting plots was significantly greater than in competition planting plots (Mann-Whitney Rank Sum test, T=334.5; n=20; P=0.042). Dense growth by species such as *B. monnieri* essentially covered plants of *S. portulacastrum* in many competition plots. There was no significant difference between final average cover of non-planted species in low-density, high-density or control plots.

<u>Conclusions</u>

a)

b)

Figure 22. *S. portulacastrum* planting plots April 2003. a) Poor health of *S. portulacastrum* in high-density no-competition plot. b) High-density "competition" plot completely overgrown with *B. monnieri* and *P. indica.*

Although *S. portulacastrum* is often found growing in areas exposed to high salinity and high temperatures. Outplantings of this species did not fare well at Honouliuli. Percent survival was 65% after a year and physical appearance in many surviving plants at this time was quite poor. After one year percent cover of *S. portulacastrum* was greater than 50% in only 6 of 40 planting plots. Initially planting *S. portulacastrum* in dense stands may increase the likelihood of establishing abundant cover of this species. During this study lower density plantings never achieved dense cover and were more rapidly overcome by non-planted species, such as, *B. monnieri* and *B. maritima.* Because *S. portulacastrum* is such a low-growing groundcover, its ability to compete with woody or bushy invasive species such as *B. maritima* and *P. indica* is limited. Without frequent vegetation control this species may be overcome by other rapidly growing shrubs and groundcovers.

The steady decline in health, survival and reproduction in *S. portulacastrum,* and several of the other study species, could also be related to a period of adjustment that plants went through after outplanting. Each plant was planted with some potting soil from the nursery. This could have resulted in the initially high survival and reproductive rates. However, as the effect of this enriched soil diminished, many plants may not have been able to adjust to the soil and local environment at Honouliuli. The plants that were able to survive and adjust to conditions at Honouliuli may have gone through a period of reduced growth and reproduction during this period. Increased health and reproduction in March and April may have occurred because these surviving plants were now acclimated to dry, hot conditions at Honouliuli. Monitoring plant populations, and especially outplanted populations, for a period of several years would be useful for answering some of these questions.

Sporobolus virginicus

a) b)

Figure 23. *S. virginicus* planting plot at (a) time of outplanting and (b) four months after outplanting.

Survival and Physical Appearance

Outplanted populations of *S. virginicus* maintained high survival rates during the entire study period. After one year, 86.8% of the original outplanted individuals were still alive. Overall survival did not drop below 90% until October 2002, six months after outplanting. As with many of the other study species, death was not randomly scattered throughout the planting plots and blocks. Fifty-two percent of final mortality was recorded from one planting block. Mortality in this area occurred rapidly after outplanting. Within one month after outplanting, only 14 of 38 *S. virginicus* plants located in this block were still alive. Only 12 of these 38 plants survived to the end of the study. Similar to other species, the block exhibiting high mortality also had the highest overall average soil salinity (12.38 mmhos; 9.90 ppt). However, the area with the second highest average soil salinity measurements exhibited 100% survival after one year and final percent survival was not correlated with overall average salinity. Final percent survival was also not related to soil temperature. Survival in *S. virginicus* appears to be related to environmental factors other than soil salinity and soil temperature. Soil substrate could have played a role, as the soil in area with high mortality appeared to contain a much higher proportion of clay than in other planting areas. Final average percent survival was not significantly different in high-density and low-density treatment plots or in competition and no-competition treatment plots.

Prior to outplanting, many *S. virginicus* seedlings were quite tall and spindly with very few leaves. In fact they hardly resembled *S. virginicus* plants found growing in the wild. Physical appearance improved dramatically within one month. Average health-code rose from 3.76 ± 0.71 at the time of outplanting to 4.79 ± 0.70 one month later. Physical appearance declined from June through November 2002, but then remained relatively stable from November 2002 through March 2003. Physical appearance improved in many of the plants after heavy rainfall which occurred in March 2003. Average health-code at the end of the study was 3.24 ± 0.90. After one year, the majority of *S. virginicus* plants were quite healthy. However plants in no-competition plots appeared more robust than plants in no-competition plots. In fact final average health-code in no-competition plots (3.67 ± 0.90) was significantly greater than in competition plots (2.78 ± 0.63) ($t = -3.564$; $df = 37$; $p = 0.001$). Final average health-code in high-density plots was not significantly different than in low-density plots.

<u>Vegetative Growth</u>

Outplanted *S. virginicus* individuals exhibited extensive vegetative growth and spread in the year following outplanting. The number of tillers per individual was estimated for *S. virginicus* because each plant produced so many tillers. The average number of tillers per individual at the time of outplanting was less than 5. The number of tillers per individual increased consistently through the study period and at the end of the study period the number of tillers per individual was greater than 15. Many individuals had produced greater than 25 tillers by the end of the study. The average maximum height of seedlings at the time of outplanting was 24.9 ± 9.2 cm. Average maximum height increased through July until it reached a maximum average of 35.4 ± 9.7 cm. After July 2002 average maximum height basically plateaued, fluctuating between 32 and 35 cm. After one year, average maximum height was 32.1 ± 7.9 cm. The average maximum height and average number of tillers was not significantly different in any of the treatment plots.

<u>Reproduction</u>

Production of reproductive stems by *S. virginicus* did not occur until June of 2002. Approximately 50% of *S. virginicus* plants were reproductive by mid-June. Approximately 80% of *S. virginicus* individuals had reproductive tillers by mid-July and the average number of reproductive stems was approximately 3.64 ± 3.96 per individual. Number of reproductive stems per individual continued to climb to its highest level, 5.05 ± 6.35, in mid-December 2002. Reproduction steadily declined from mid-December through the end of the study. By April 2003, less than 10% of *S. virginicus* plants were reproductive and average number of reproductive stems per individual had dropped to 0.39 ± 1.72. The actual amount of viable seed produced by *S. virginicus,* however, is questionable. When flowering stalks were examined, if seeds existed they often appeared white, indicating that viable seeds were not being produced by all reproductive stems.

Final average number of reproductive stems was similar in all density and competition treatments.

<u>Percent Cover</u>

Initial percent cover of *S. virginicus* in outplanting plots averaged 31.4 ± 3.8 in high-density and 19 ± 5.0 in low-density plots. Despite slightly declining health and increasing mortality, percent cover of *S. virginicus* continued to rise throughout the study period. This is indicative of the continuous vegetative growth and vegetative spread of *S. virginicus* throughout the study period. Dense cover was provided by both high-density and low-density plots. In fact, by August 2002 the average percent cover in low-density plots (54.0 ± 19.5) was similar to average cover in high-density plots (59.6 ± 21.6). By April 2003, one year post-outplanting, average percent cover of *S. virginicus* had risen to 73.7 ± 26.2 in high-density, 66.9 ± 27.5 in low-density and 14 ± 21.4 in control plots. There was no significant difference between final average percent cover of low vs. high-density plots. But final average percent cover in no-competition plots (82.0 ± 17.3) was significantly greater than in competition (58.6 ± 29.6) plots (t = 3.049; df = 38; p=0.004). Colonization of non-planted species appears to have reduced the vegetative cover of *S. virginicus* in competition plots. Plants in no-competition plots were able to spread more readily resulting in higher final average percent cover.

a)

b)

Figure 24. April 2003: a) low-density / competition plot being overgrown by *P. indica;* b) low-density/ no-competition plot.

A one-way ANOVA showed a significant difference among competition treatment plots in final average percent cover of non-planted species ($F_{2,27}$=6.387, p=0.005). Tukey's HSD identified that average percent cover of all non-planted species in control plots (154.2 ± 34.16) was significantly greater than in low-density (105 ± 45.71; p=0.036) or high-density (90.4±44.63; p=0.006) plots. Apparently, the dense cover of *S. virginicus* reduced the ability of non-planted species to colonize planting plots.

Conclusions

Outplanted *S. virginicus* populations had the second highest survival rate of all outplanted species. Plants survived and flourished in almost all planting plots at Honouliuli. Vegetative spread exhibited by *S. virginicus* was quite extensive. In fact *S. virginicus* plants had spread to 15 of 20 control plots by the end of the study period. As with other species, no seedlings were observed during the study period and the viability of seeds produced by flowering stems was questionable. Spread of *S. virginicus* at Honouliuli will most likely continue through vegetative means versus through seed dispersal.

Dense cover was quickly provided by *S. virginicus* plants in both high and low-density planting plots. Higher density outplantings of *S. virginicus* may not be necessary. During this study lower-density planting plots provided cover as abundant as high-density planting plots within four months. However, percent cover after one year was significantly greater in no-competition plots than in competition plots, thus in order to maintain abundant cover of *S. virginicus* periodic vegetation control of non-desired species should be undertaken.

IV. Results and Conclusions: Germination Trials

Three germination trials to investigate testing germination response under different water-levels, salinities, and temperatures on germination of native sedge species were conducted between August 2002 and April 2003. Fresh seeds of *B. maritimus, C. javanicus, C. laevigatus,* and *C. polystachyos* were used for each trial. Five replicates of twenty seeds per species and per treatment were tested during each trial. Seeds were germinated in Petri dishes on Whatman No. 1 filter paper. Petri dishes with tight fitting lids were used to reduce water loss. Water levels were checked and germinated seeds were removed every other day. Distilled water was used for all treatments. Seeds were placed in temperature controlled growth chambers at the University of Hawai`i at Manoa for the duration of each trial. Each trial lasted for approximately eight weeks.

Percent germination data were arcsine-square-root transformed prior to analysis. Data was then analyzed for statistical significance using a one-way ANOVA. Multiple comparisons of means were made using Tukey's test of 'honestly significant differences'. All tests of significance were conducted at the $p<0.05$ level. Percent germination in the dry water-level treatment was 0% for all species in all temperature regimes, thus, this treatment is excluded from data analysis. All other treatments that resulted in 0% germination were also not included in data analysis.

Results: Trial 1: August 2002 – October 2002

The initial test of seed germinability was conducted from August to October 2002. This trial investigated germination response under a range of water levels. Five replicates of twenty seeds of each species were placed under four water-levels: 1 cm of standing water, saturated (water just covering the top of the seeds), moist (moistened filter paper) and dry. Seeds were placed in temperature controlled growth chambers with an alternating temperature regime of 29/23°C and 12 hour light/dark photoperiods with light corresponding with the higher temperature. The temperatures were chosen based upon the average monthly temperature for southwest Oahu from July through October.

After eight weeks, there was very low germination in any of the species except for *C. polystachyos.* Final germination percent for *C. polystachyos* was highest under moist conditions (94%) followed by saturated conditions (64%), and 1 cm of water (40%). No seeds germinated under dry conditions (Fig. 25). A one-way ANOVA of final germination percentages showed a significant effect of water level on overall germination percentage ($F=129.611$; $p<0.001$). Germination under moist conditions was significantly greater than germination under either saturated conditions or 1cm of standing water ($p<0.001$ for both combinations). Germination under saturated conditions was also significantly greater than germination percentage under 1cm of standing water ($p=0.008$). Seeds of all other species showed very low germination with percentages between 0 and 6%, under all moisture levels. Germination percentage in these species was too low to analyze for differences due to water-level treatment.

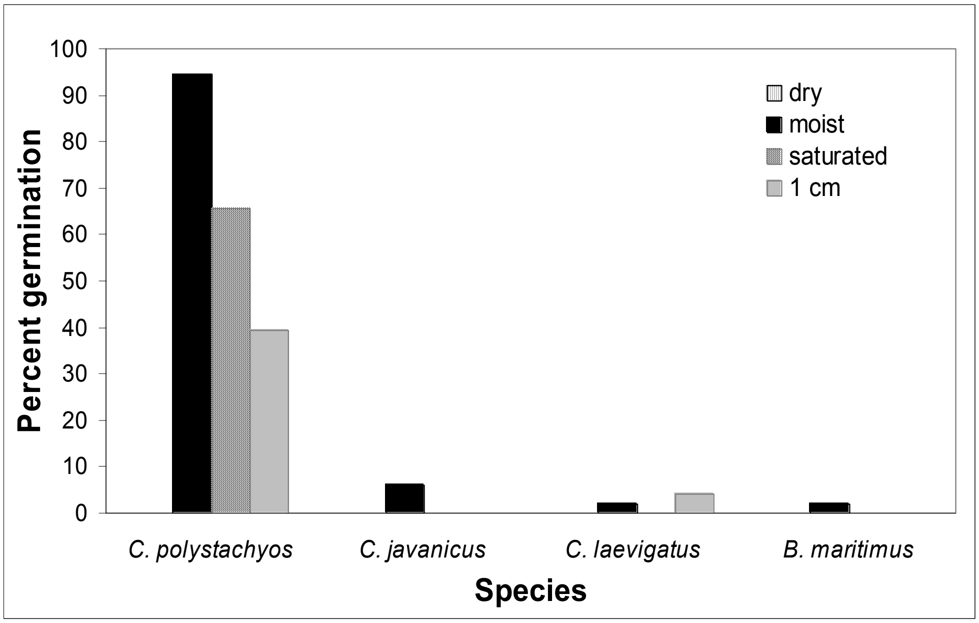


Figure 25. Final percent germination of native wetland sedges after eight weeks in four water-level treatments under an alternating light (12 hour dark/light) and temperature regime (24.3/18.2°C).

Results: Trial 2: October 2002 to December 2002

In October 2002 a second germination experiment was conducted. This trial tested germination under different water levels and salinity conditions. Five replicates of twenty seeds for each species were placed under four different levels of salinity: 0, 8.5, 17, and 34 parts per thousand (ppt). Salinity solutions were prepared by diluting filtered sea water with distilled water to the correct ratio. Water-level treatments were as described above. Based on the lack of results from the first trial, seeds were placed in a growth chamber with an alternating 24.3:18.2 °C (versus 29/23°C) temperature regime with 12-hour alternating light/dark photoperiods.

Final germination percentages for water-level treatments ranged from 0%, for seeds of *B. maritimus* and *C. laevigatus* under all treatments, to 85% for seeds of *C. javanicus* in the saturated treatment (Fig. 26). Final germination percentages for *C. javanicus* was highest under saturated water-level (85%), followed by moist conditions (78%), and 1cm standing water (55%). A one-way ANOVA of final germination percentages showed a significant effect of water level on overall germination percentage of *C. javanicus* ($F=6.61$; $p=0.01$). Final germination percentage under saturated conditions was significantly greater than under one cm of standing water ($p = 0.01$). Germination under moist conditions was not significantly different than germination under saturated conditions.

Final germination percents for *C. polystachyos* ranged from 65% under one cm of standing water to 78% under saturated conditions. Final germination in moist soil conditions (76%) was only slightly lower than under saturated conditions. There were no significant differences in final germination percentage of *C. polystachyos* in the water-level treatments.

a) b)

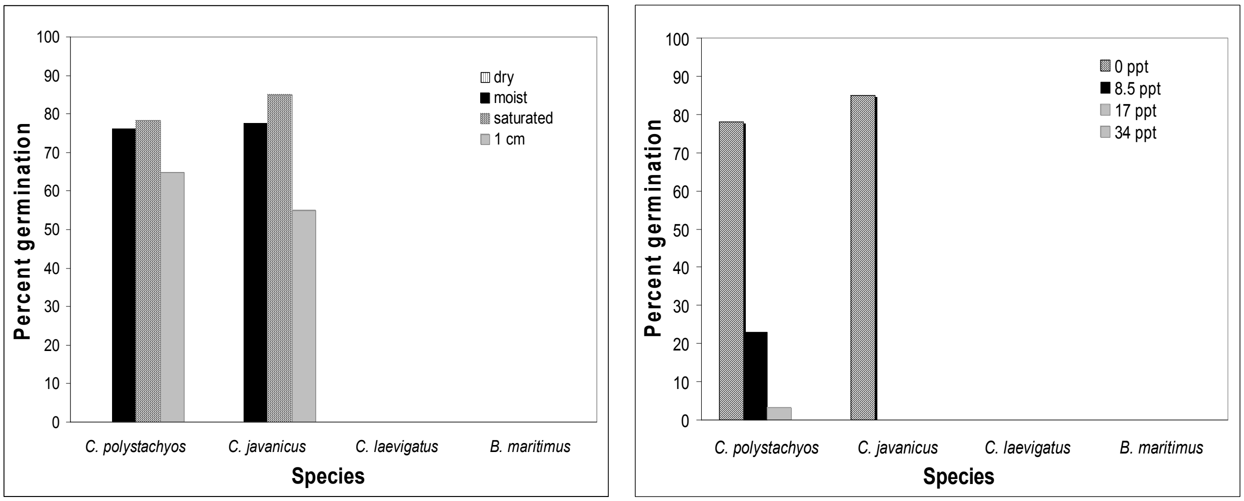


Figure 26. Final percent germination of native wetland sedges after eight weeks in (a) four water-level treatments and (b) four salinity treatments under an alternating light (12 hour dark/light) and temperature regime (24.3/18.2°C).

Seeds of all species, except *C. polystachyos,* failed to germinate under salinity conditions of 8.5, 17, and 34 ppt. Final germination for *C. polystachyos* was 23% under a salinity of 8.5 ppt, 3% under a salinity of 17 ppt, 0% under 34 ppt and 78% in freshwater (0 ppt).   A one-way ANOVA of final germination percentages showed a significant effect of salinity level on overall germination percentage of *C. polystachyos* ($F=48.754$; $p<0.001$). Final germination percentage was significantly greater in the 0 ppt treatment than in either the 8.5 ppt or 17 ppt treatments ($p<0.001$ for both combinations). Germination was also significantly greater in the 8.5 ppt treatment than in the 17 ppt salinity treatment ($p=0.005$).

Results: Trial 3: February to April 2003

A final experiment testing germination response under different water-levels, salinities and temperatures was conducted beginning in February 2003. The results of the first two trials demonstrated that temperature has a distinct influence on germination of, at least, *C. polystachyos* and *C. javanicus*. Thus, a 4 x 3 factorial experiment was performed with four water-levels (1cm standing water, saturated, moist, dry) and three alternating temperature regimes (29/23, 24.3/18.2, and 20/15 °C). Concurrently, a 4 x 3 factorial experiment was conducted with four salinity levels (0, 8.5, 17, and 34 ppt). As in previous trials, five replicates of twenty seeds for each species and treatment were placed in growth chambers with alternating temperature regimes and 12 hour light/dark photoperiods. Water levels and saline solutions were checked and germinated seeds were removed at two-day intervals for eight weeks. Due to lack of any germination in previous trials, seeds of *B. maritimus* were scarified by soaking them in a bleach solution (3% sodium hypochlorite) for three days (following Clevering 1995) prior to being placed under treatment conditions.

Germination results under the various treatment conditions are summarized in Table 4. Similar to the results of trial one and trial two, percent germination of *B. maritimus* and *C. laevigatus* was very low under all treatments and temperature regimes. However, unlike the first two trials, in which *B. maritimus* germination was 0% in almost all treatments, seeds of *B. maritimus* did germinate under several treatments in trial three. This is presumably due to the bleach scarification treatment. Overall, seeds of *B. maritimus* germinated to a greater extent under the 24.3/18.2 ° C temperature regime than in the 29/23 or 20/15 °C temperature regimes. Final germination percents were significantly greater in the 24.3/18.2 than in the 29/23° C temperature regime (mean percent germination: 19.0 ± 11.2 in 24.3/18.2 °C and 10.3 ± 6.1 in 29/23°C; $t = 2.628$; $df=28$; $p=0.014$). No germination of *B. maritimus* was observed in any treatment in the 20/15 °C temperature regime, thus this temperature regime was not included in statistical analysis.

Maximum germination (24%) for *B. maritimus* occurred under conditions of 1 cm of standing water at 24.3/18.2 °C (Table 4; Fig. 27d). Regardless of temperature regime, seeds of *B. maritimus* did not germinate in salinities greater than 0 ppt. There were no significant differences due to water-level treatments in either the 24.3/18.2 °C or the 29/23 °C temperature regimes.

Table 4. Mean percent germination after eight weeks for seeds of native species under different water-levels, salinity levels, and temperature regimes.

Species/ temperature	**Water Level**				**Salinity**			
C. polystachyos	**dry**	**moist**	**saturated**	**1 cm**	**0 ppt**	**8.5 ppt**	**17 ppt**	**34 ppt**
20/15 °C	0%	88%	95%	82%	95%	60%	0%	0%
24.3/18.2 °C	0%	98%	82%	84%	82%	30%	0%	0%
29/23 °C	0%	74%	78%	68%	78%	26%	0%	0%
C. javanicus								
20/15 °C	0%	80%	90%	87%	90%	70%	0%	0%
24.3/18.2 °C	0%	90%	80%	88%	80%	74%	0%	0%
29/23 °C	0%	46%	24%	28%	24%	1%	0%	0%
C. laevigatus								
20/15 °C	0%	2%	3%	7%	3%	0%	0%	0%
24.3/18.2 °C	0%	0%	1%	0%	1%	0%	0%	0%
29/23 °C	0%	0%	3%	8%	9%	0%	0%	0%
B. maritimus								
20/15 °C	0%	0%	0%	0%	0%	0%	0%	0%
24.3/18.2 °C	0%	17%	16%	24%	16%	0%	0%	0%
29/23 °C	0%	12%	8%	11%	8%	0%	0%	0%

Average percent germination for both *C. javanicus* and *C. polystachyos* was significantly higher under alternating temperatures of 20/15 and 24.2/18.3°C than 29/23 °C [*C. javanicus:* F=158.44 $p<0.001$; $p<0.001$ for both 20/15 and 24.3/18.2 °C vs. 29/23°C. *C. polystachyos:* F=8.10; p=0.001; p=0.003 for both 20/15 and 24.3/18.2 °C vs. 29/23°C]. There was no overall effect of water level on germination for either *C. javanicus* or *C. polystachyos.* Final percent germination for *C. javanicus* was highest under moist conditions at 29/23 and 24.3/18.2 °C, but was lowest under this condition at 20/15 °C (Table 4; Fig. 27). Similarly, mean germination percent was highest under saturated conditions for seeds of *C. polystachyos* at 20/15 and 29/23 °C, but was lowest under saturated conditions at 24.3/18.2 °C.

Final germination percent for *C. javanicus* in the 29/23 °C temperature regime was significantly greater under moist conditions than either saturated or 1 cm standing water at (F=16.05, $p<0.001$) ($p<0.001$ and p=0.008 for most vs. saturated and moist vs. 1cm respectively). Percent germination was not significantly different for *C. javanicus* seeds under different water-level conditions in the 24.3/18.2 and 20/15 °C temperature regimes.

Final percent germination was also significantly greater in moist conditions than in saturated or 1cm standing water conditions for seeds of *C. polystachyos* at 24.3/18.2 °C (F=10.925, p=0.002; moist vs. saturated, p=.004; moist vs. 1 cm., p=0.005). Final germination percents of *C. polystachyos* were not significantly different in the water-level treatments in the 29/23 and 20/15 °C temperature regimes.

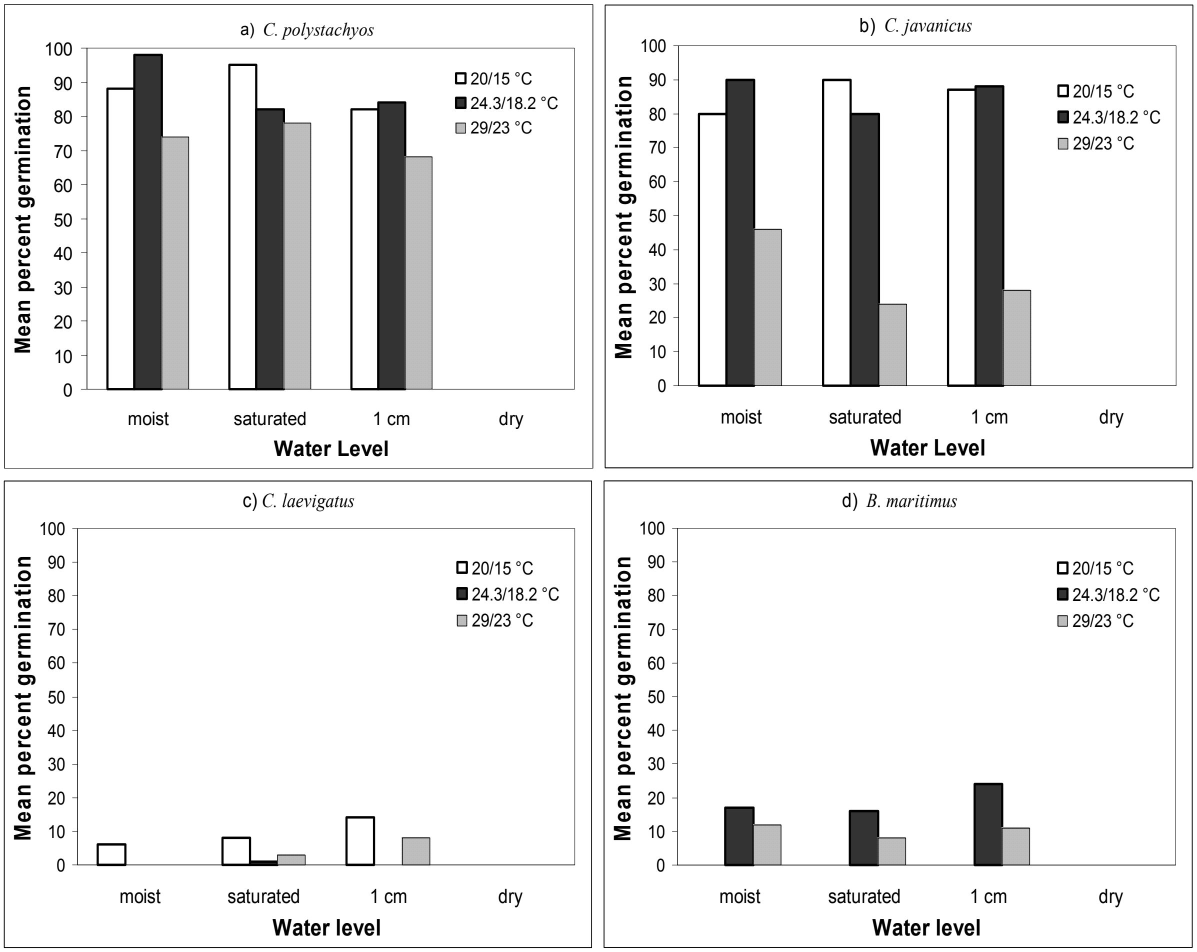


Figure 27. Mean final percent germination of native wetland sedges after eight weeks under different water-levels, salinity and temperature regimes.

Conclusions

Of the four species studied, only *C. polystachyos* and *C. javanicus* exhibited any substantial germination under experimental treatment conditions. *C. polystachyos* appeared able to germinate readily in a wide variety of temperatures and water-levels. *C. polystachyos* also showed moderate levels of germination under slightly saline conditions. Further, although seeds of *C. polystachyos* did not germinate under higher levels of salinity, when seeds stored in saline conditions were given fresh water they rapidly germinated. Sowing seeds of *C. polystachyos* in addition to, or even instead of, transplanting seedlings may be a viable restoration option for this species.

Despite the fact that *C. javanicus* is found growing in areas that experience high average temperatures, seeds of *C. javanicus* did not readily germinate at higher temperatures. Germination occurred to a greater extent under fluctuating temperatures of 24.3/18.2 and 20/15 °C than when subjected to alternating temperatures of 29/23 °C. This implies that seeds of *C. javanicus* may germinate in the winter or spring when average temperatures are lower. Thus, if seeds are to be used in a restoration effort, it may be wise to either sow seeds in early winter or early spring. Seeds of *C. javanicus,* under lower temperatures, germinated under all water-levels. Sowing seeds in an area that may experience shallow flooding or waterlogged soils may not hinder germination of *C. javanicus.* As with *C. polystachyos,* seeds of *C. javanicus* only germinated under low salinity levels. However, when seeds, which were previously exposed to higher salinity levels (17 ppt and 34 ppt),were placed in fresh water they exhibited high germination percents.

Seeds of both *B. maritimus* and *C. laevigatus* showed low germination under all temperatures, water-levels and salinities. *B. maritimus* seeds have a hard seed coat and it appears that they must break this physical dormancy in order to germinate. Bleach-scarification increased the germination rates in this species, but even so, final germination percent of bleach-scarified seeds never exceeded 25% in any of the experimental conditions. Mechanically scarifying *B. maritimus* seeds may increase germination of this species. Unlike *C. polystachyos* and *C. javanicus,* bleach-scarified seeds of *B. maritimus* did not germinate under the lowest temperature regime (20/15°C). Germination requirements of and techniques for breaking dormancy in this species should be further tested prior to using seeds to restore or augment populations of *B. maritimus.*

In all three trials, final germination of *C. laevigatus* never exceeded 10% in any of the temperature, water-level, or salinity conditions. Initially, seeds of *C. laevigatus* were presumed unviable. However, by placing seeds of *C. laevigatus* in colder temperatures (20/15°C) for several weeks and then placing them under a higher temperature regime (29/23 °C) relatively high rates of germination were achieved, especially under saturated and 1cm of standing water. Results of these "trials" have not been included in this report as they were not obtained from controlled studies. But, it does present an intriguing area for further investigation into germination requirements of this species. Other researchers have been able to achieve high rates of *C. laevigatus* germination in nursery settings, but the conditions under which germinating seeds were exposed have not been documented (Van Dyke, pers. comm.) This species is readily propagated through divisions and this method would be recommended for restoration efforts until further knowledge of germination requirements of this species are understood.

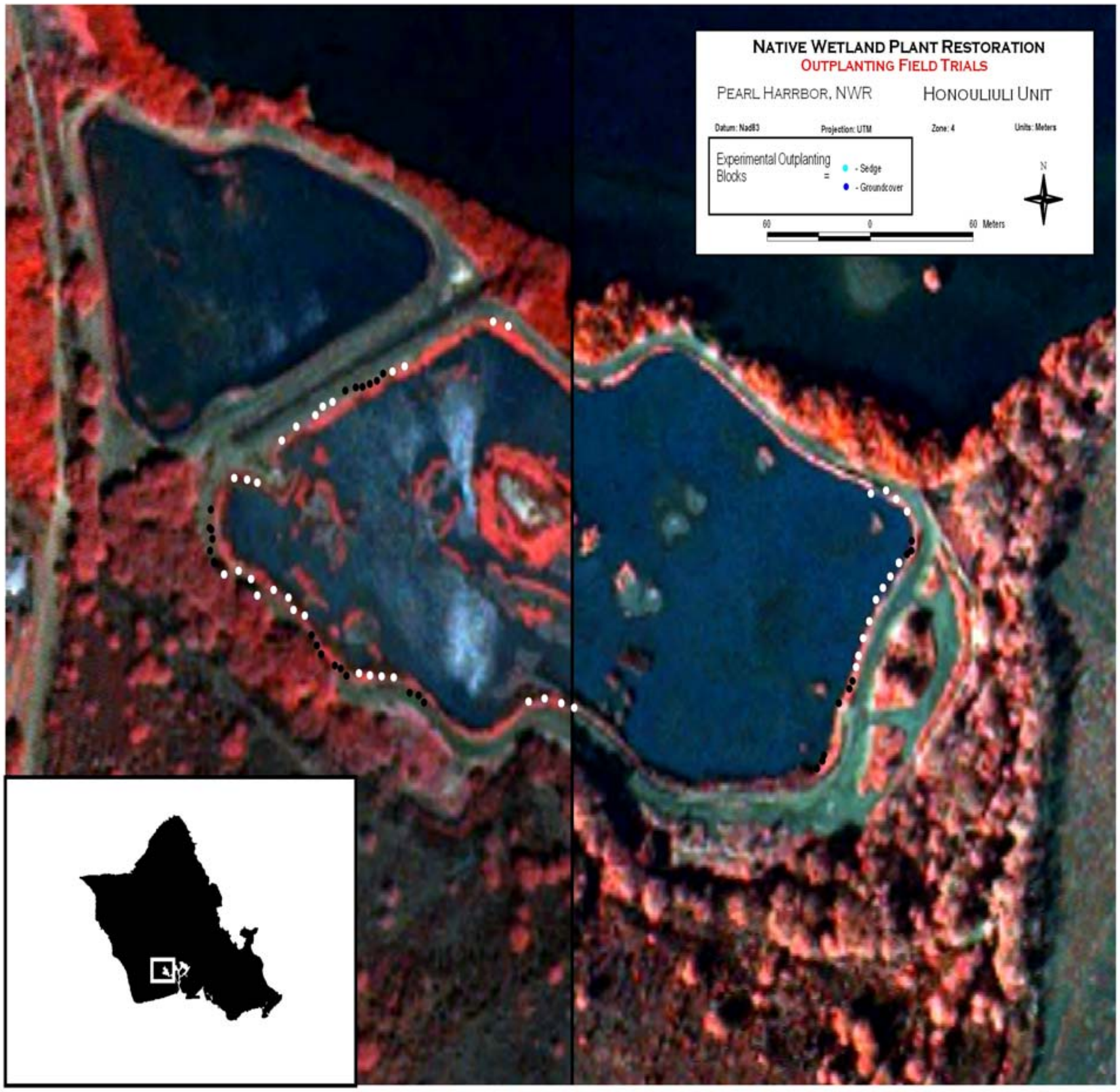
Germination requirements of native wetland species needs to be studied further. In these experiments, only fresh seeds were used. In many restoration efforts fresh seeds may not be readily available. Thus, studying germination response after different lengths of storage or different storage regimes would be useful. Further, many seeds of species in these studies were relatively slow to germinate. Higher germination percents could possibly be exhibited if seeds are allowed to germinate for greater than eight weeks. Lastly, germination phenology studies in the field, for example recording when seeds are set and when seeds germinate in natural populations, would greatly increase out knowledge of germination requirements and would add to our scant knowledge of native wetland species.

IV. Results and Conclusions: Attractiveness of Native Plant Species to Native Waterbirds

Because the native plant outplanting plots were relatively small and scattered around the pond, native waterbirds were rarely if ever observed in these areas. However, nests of Hawaiian coot (*Fulica americana alai)* were often observed in natural populations of *B. maritimus* at Honouliuli (pers. obs.; Silbernagle, pers. comm.). Individuals of *F. Americana alai* and Hawaiian moorhens (*Gallinula chloropus sandwicensis)* have also nested in stands of *B. maritimus* and *C. javanicus* (pers. obs.; Silbernagle, pers. comm. 2001). *F. Americana alai* individuals have also been observed feeding on *B. maritimus, C. javanicus,* and *C. polystachyos* seed heads and leaves of *B. maritimus* and *C. polystachyos* (pers. obs.; Silbernagle, pers. comm. 2001). *F. Americana alai* and *G. chloropus sandwicensis* presumably feed on seed heads of *C. laevigatus* and use stems of *C. laevigatus* in other wetlands around the state; however they have not been observed doing so at Honouliuli.

Endangered Hawaiian stilts (*Himantopus mexicanus knudseni*) use little vegetation for nesting or feeding. They have a tendency to nest on bare soil and feed primarily on invertebrates (USFWS 1985). *H. mexicanus knudseni* individuals were often observed feeding on invertebrates in mats of *B.monnieri* on the edge of Pond 2 at Honouliuli (pers. obs.). *B. monnieri*, though not one of the outplanted species, is a native groundcover found in large natural populations at Honouliuli.

Appendix 1: Experimental Outplanting Field Trial Plot Locations



○ = 1.75 x 6.5 m sedge plots; ● = 2 x 3.5 m groundcover plots

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Appendix 2: Annual Report: June 3, 2002

ANNUAL REPORT:
Research on Native Plants for Coastal Wetland Restoration on O'ahu
June 3, 2002

Agreement 12200-1-J005

To

The U.S Fish and Wildlife Service
Oahu, Hawaii

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Project Researcher
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University of Hawaii
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This annual report covers work completed from August 2001 through May of 2002.

I. Outer Island travel

Kona Coast – Island of Hawai'i

In August of 2001 I explored two important wetlands on the Kona coast with Kim Uyehara of Ducks Unlimited. The two wetlands visited were Makalawena (Opaeula Pond) and Kaloko-Honokohau National Park.

Located on Kamehameha Schools/Bishop Estate land, Opaeula Pond is a coastal wetland adjacent to a series of anchialine pools. Opaeula Pond is an important waterbird sanctuary. The salinity of this pond averages between 4-10 parts per thousand (ppt) (Uyehara 2001, personal communication). A large variety of native plant species grow in and around Opaeula Pond. I noticed that makaloa (*Cyperus laevigatus)* was fairly abundant and was often seen growing just inland of stands of bulrush (*Scirpus* spp*.)*. Makaloa also appeared in areas with little soil or water, even growing out of lava rock. Makai (*Bolboschoenus maritimus)* is another native sedge that is quite abundant at Opaeula Pond. Makai and makaloa often formed interspersed stands around the edges of the pond. Other native plants seen in abundance at Opaeula Pond include: \`ae \`ae (*Bacopa monnieri*, ohelo kai (*Lycium sandwicense*) and \`akulikuli (*Sesuvium portulacastrum)*.

After touring Opaeula Pond we visited the wetlands at Kaloko-Honokohau National Park. This wetland is surrounded on three sides by a relatively recent lava flow. The fourth side fronts the Pacific Ocean. Unfortunately this wetland does not support the abundant native vegetation seen at Opaeula Pond. Once again it was noticed that lava does not preclude wetland vegetation as invasive pickleweed (*Batis maritimus)* was seen growing right in the lava rock. Unfortunately, *B. maritimus* is not the only invasive species in abundance at Kaloko-Honokohau. Also abundant at Kaloko-Honokohau is salt-grass (*Paspalum distichum),* a non-native, invasive grass that grows in many wetlands around the state. Salt-grass grows out from the edges of the pond right into the open water. Control efforts for salt-grass is seen as necessary to open up stilt nesting habitat and allow for the growth of native wetland plants, such as makai (*Bolboschoenus maritimus)* (Uyehara 2001, pers. comm.). Makai grows only sporadically at Kaloko-Honokohau and must compete with the abundant *P. distichum*. Other native species at Kaloko-Honokohau include naupaka (*Scaevola sericea)* and akulikuli (*Sesuvium portulacastrum)*.

While visiting the Kona coast wetlands Kim Uyehara also discussed Ducks Unlimited involvement with mid-to-upper elevation wetlands on the Island of Hawaii. This work involves not only learning how to control invasive plant species but also trying to create habitat for native damselflies. In the future they may focus more on outplanting of native plant species as they have found that not enough emergent vegetation has come up in some of these upper elevation ponds. Work with upper elevation ponds focuses mostly on benefits to Koloa ducks and nene.

Kauai

In December of 2001 one day was spent day touring Kauai wetlands with Adam Asquith, Kauai's SeaGrant Extension Agent. We began by visiting Kawaiele Bird Sanctuary. Formerly the site of a sand mining operation, this area became a sanctuary when the water table was reached and the area flooded. The area originally attracted a large number of waterbirds including the Hawaiian moorhen (*Gallinula chloropus sandvicensis*) and Hawaiian coot (*Fulica americana alai)*. However, soon after creation of the sanctuary, tilapia fish (*Tilapia* spp.) were introduced into the pond. *Tilapia* spp. are omnivorous with voracious appetites. The introduction of tilapia drastically decreased the productivity of the ponds. Native and migratory waterbirds still visit the ponds, but they don't tend to stay (Asquith 2001, pers. comm.). Currently, this area is overrun by invasive plant species and native plants are not found in the sanctuary. Possibly, this is due to the fact that this is a created wetland. Historically this area may have been part of a larger dune system, thus a native wetland seedbank probably does not exist in this area (Asquith 2001, pers. comm.).

Next we headed east toward Hanapepe. Much of the area between Waimea and Hanapepe consists of a flat plain between the ocean and the mountains. Historically, this area was probably characterized by an extensive coastal dune system with vast seasonal wetlands forming in the depression between the ocean dunes and the mountain face (Asquith 2001, pers. comm.). Asquith believes that this area originally would have supported sedges and smaller shrubs but not trees. Next we stopped by the Hanapepe river mouth around Hanapepe bay. Currently, this area is overrun by pickleweed (*Batis maritima)* and Indian fleabane (*Pluchea indica)*.

After leaving Hanapepe we headed North towards Hanalei. On our way to Hanalei we stopped outside Kapaa at Nukoli'i pond. This small pond was also the site of a former sand mining operation. Soon after sand mining was halted, native plants including 'ae 'ae (*Bacopa monnieri)* and makai (*Bolboschoenus maritimus)* rapidly began to re-vegetate this area.

Hanalei National Wildlife Refuge was our next stop. We toured the ponds at Hanalei and discussed Adam Asquith and Christian Melgar's recent study on the response of native waterbirds to pond water depth and disturbance regimes. Their study utilized 12 replicate ponds with different water depths and disturbance frequencies. "Disturbance" consisted of mowing down all existing vegetation. Results of their study indicated that shallow water (2-9 cm) supported more waterbird and plant diversity than either deep water (over 17 cm) or sheet-flow water regimes. Interestingly they also noticed that even replicate ponds with the same water depth and disturbance regime supported a heterogeneous assortment of plant species. Following disturbance, native species, such as sawgrass (*Cladium jamaicense),* `ahu `awa (*Cyperus javanicus)* and great bulrush (*Schoenoplectus lacustris),* germinated from seed and persisted in areas of shallow water.

Wetland Delineation Training – Kauai

In January of 2002 I attended a wetland delineation workshop offered by the Natural Resources Conservation Service (NRCS). The training, held in Kapaa, Kauai, was an intensive two-day training course. The first day was spent in the classroom studying how hydrology, soils, and vegetation characteristics are used to delineate wetlands. The next day was spent out in the field. In the field we broke in to teams and practiced using our classroom learning in a real-life setting.

The workshop was an incredibly valuable workshop, and taught me the intricacies and difficulties with determining if an area, especially an area that has been heavily altered, exhibits true wetland characteristics.

II. Restoration of wetlands in Hawai'i

A literature search, as well as extensive personal communication with wetland managers and researchers around the state, provided the framework for the first report submitted to the USFWS (See Appendix I: Wetland Restoration in Hawaii). This report summarized the scant literature regarding wetland restoration in Hawai'i. This review also summarized paleo-botanical literature and discussed the value of this type of work to wetland restoration in Hawai'i. The reality uncovered through writing of this review is that wetland restoration in Hawai'i is quite a new endeavor and rarely are efforts at restoration documented and published. To date most wetland "restoration" in Hawai'i has simply involved removal of non-native, invasive vegetation. Little effort has been focused on actively outplanting native wetland vegetation. This review establishes that learning how to effectively restore native plants to Hawaii's wetlands will require much more effort, scientific study and most importantly documentation of successes and failures encountered by those attempting to restore wetland vegetation.

III. Research proposal

The attached research proposal (See Appendix II) outlines the scope of work to be completed through field and lab studies.

IV. Field research

In August of 2001, I began researching potential native plant species to be utilized for wetland outplanting field trials. The list was narrowed down to eight plant species, five sedges and three groundcovers. Next began the search for a nursery or grower familiar with propagation and cultivation of native wetland plants. I soon realized that the number of nurseries in the state growing native plants is quite small and rarely do these nurseries cultivate wetland plants. The Research Corporation of the University of Hawaii (RCUH) requires that three price quotes be obtained prior to purchases of greater than $2500. Thus, the process of finding growers and receiving price quotes for cultivation of a large number of wetland plant species became a lengthy process. By the time all three required price quotes had been obtained, a grower had been chosen and a purchase order from RCUH had been received it was well into October of 2001.

The lowest price quote came from Hui Ku Maoli Ola Native Hawaiian Plant Nursery. I collected and supplied seeds of four of the eight plant species to Hui Ku Maoli Ola. Seeds of four of the five sedge species, \`ahu \`awa (*Cyperus javanicus),* aka \`akai (*Schoenoplectus lacustris),* makai (*Bolboschoenus maritimus)* and *Cyperus polystachyos,* were collected from Pearl Harbor and James Campbell National Wildlife Refuges in September of 2001. The other four plant species: makaloa (*Cyperus laevigatus),* pa\`uohi\`iaka (*Jacquemontia ovalifolia),* \`akulikuli (*Sesuvium portulacastrum)* and \`aki\`aki (*Sporobolus virginicus)* either do not grow at James Campbell and

Pearl Harbor NWR or are more readily propagated through cuttings. Hui Ku Maoli Ola supplied the source for these remaining species.

The owners of Hui Ku Maoli Ola estimated that it would require roughly three months to grow approximately 300 - 400 plants of each of the eight plant species listed above. Therefore, they estimated that the plants would be ready for outplanting in mid-January. Unfortunately by mid-January, due to slow germination and perhaps the incessant rains of the winter of 2001/2002, the seedlings were still too small to outplant. Further, despite several attempts, the growers at Hui Ku Maoli Ola were unable to germinate seeds from aka`akai (*S. lacustris)* or makai (*B. maritimus)*.  I collected several batches of seed from Pearl Harbor NWR as well as at Hamakua Marsh (for the aka`akai) and none of the batches of seeds germinated. We agreed to forgo further attempts to try and propagate these two species. I decided that I would not attempt to use *S. lacustris* for the outplanting trials and that I would transplant individuals of *B. maritimus* from existing populations at Honouliuli Unit of Pearl Harbor NWR. February of 2001 rolled around and the plants were still too small to successfully outplant. Finally in March of 2002, the growers and I decided that outplanting could begin.

Prior to outplanting, the field sites needed to be cleared of vegetation. In February of 2002 it was decided that outplanting efforts would be focused solely at Pearl Harbor NWR. Outplanting plots were selected from around the edge of Pond 2 at the Honouliuli Unit of Pearl Harbor NWR (see Appendix III: Map of Honouliuli Unit, Pearl Harbor NWR). From March 9-16 forty blocks of approximately 8.125 m^2 and thirty blocks of approximately 8.75 m^2 were manually cleared of all vegetation (see Appendix II: Research Proposal for specifics on experimental design) in early March of 2002. Much of this vegetation consisted of the invasive alien species pickleweed (*Batis maritima)* and Indian fleabane (*Pluchea indica).*

After the existing vegetation was removed, outplanting was initiated. Three hundred *C. polystachyos,* 260 `ahu `awa (*C. javanicus),* 300 `akulikuli (*S. portulacastrum)* and 380 `aki`aki (*S. virginicus)* seedlings were outplanted the week of March 16 – March 23, 2002.  The following week, March 24 – March 31, 300 makaloa (*C. laevigatus)* seedlings and 260 makai (*B. maritimus)* transplants were outplanted.  Due to nursery error, the 300 pa`uohi`iaka (*J. ovalifolia)* seedlings were not ready for outplanting until early May of 2002. Plants have been monitored every two weeks since outplanting (see Appendix II: Research Proposal for specifics on monitoring protocol). Monitoring of abiotic factors of soil moisture, soil salinity and soil temperature has, at this point, been sporadic due to equipment failure. The soil salinity, soil moisture and temperature meter that has been utilized for this project has continuously malfunctioned. We, Mike Sibernagle of the Oahu National Wildlife Refuges and I, have been in continual contact with the company (Aquaterr) supplying and repairing this meter. They have assured us that a working meter will be supplied to us by May 31, 2002.

V. Conclusions to date

Of the original 2,100 seedlings/transplants outplanted, currently 2,048 were still alive as of May 20, 2002. The plants have thrived, possibly in part due to the mild and wet conditions that have continued through May of 2002. Amazingly enough, the seedlings rapidly acclimated to their new environment at Pearl Harbor NWR. In fact, within two weeks of outplanting, the majority of the plants appeared much healthier, stouter and with richer color, than when received from the nursery.

By May 20th of 2002, at least a few individuals of all species have flowered and many have set seed. As well, all species, with the exception of pa`uohi`iaka (*J. ovalifolia)* and `akulikuli (*Sesuvium portulacastrum),* have reproduced vegetatively by sending out new shoots from underground rhizomes or tubers.

Another interesting observation concerns the composition of non-planted species that have returned in each plot. To date, it has been mainly native species, primarily `ae`ae (*Bacopa monnieri)* and makai (*B. maritimus)* that have appeared in each planting plot. In all sedge plots combined, 85.7% of the cover of non-planted species has been comprised of `ae`ae (*B. monnieri).* In the groundcover plots, makai (*B. maritimus)* has accounted for 27.4% of the overall cover of non-planted species. Another native species that has showed up in many of the planting plots is kipukai (*Heliotropium curassavicum).* Clearly native species have responded well to the removal of invasive plant species such as Indian fleabane (*Pluchea indica)* and pickleweed (*Batis maritima).* It will be interesting to assess if and when these native plant species will be displaced by the inevitable encroachment of aggressive *P. indica* and *B. maritima.*

The following section provides a species by species account of the performance to date of outplanted wetland species.

`Ahu `awa (*Cyperus javanicus)*

Two hundred and sixty *C. javanicus* seedlings were planted on March 16-17, 2002. Outplanted seedlings had, on average, 5.5 tillers and an average maximum leaf length of 67.5 cm. By April 23, 2002, a few plants had started to send out reproductive shoots. By May 15th, each outplanted individual consisted of an average of 8.7 tillers and an average of 0.3 reproductive shoots. Only one *C. javanicus* seedling had died as of May 15, 2002. Overall, the outplanting of *C. javanicus* appears quite successful at this point. Currently, the plants seem quite drought tolerant and have high survival and reproductive rates. Both the high and low-density plots (see methods in Appendix II: Research Proposal) have filled in quite nicely and have been able to retard some encroachment of other plant species at this point.

Makai (*Bolboschoenus maritimus)*

Despite the fact that *B. maritimus* grows quite abundantly at the Honouliuli Unit Pearl Harbor NWR, the growers at Hui Ku Maoli Ola were unable to germinate freshly collected seeds of this species. Future germination trials (See Appendix II: Research Proposal) may help narrow down the germination requirements of *B. maritimus* seeds.

On March 24th and 25th of 2002, two hundred and sixty *B. maritimus* plants were culled from existing populations at Pearl Harbor NWR and transplanted into study plots. Transplants, on average, consisted of 1.06 shoots (all vegetative) with a maximum height of 21.4 cm. By May 15th of 2002 eight of the original 260 transplanted individuals had died. This amounts to only 3.1% of transplanted individuals. Further, by May 15th, the average number of shoots per transplant had increased to 3.5 and the average maximum height had increased to 37.9 cm. This is an increase of 77% in just over one and a half months. By May 15th 165 of the living 252 *B. maritimus* transplants contained at least one reproductive shoot. Overall, the average number of reproductive shoots for all individuals was 1.2.

Similar to *C. javanicus,* makai plants (*B. maritimus)* transplants have performed quite well. However, by the end of May, 2002 many of the individual shoots of the transplanted plants had reached the end of their reproductive phase and were beginning to senesce. It will be interesting to see if the underground tubers of the transplanted individuals continue to send up new shoots.

Makaloa (*Cyperus laevigatus*)

Three hundred *C. laevigatus* seedlings were outplanted at Pearl Harbor NWR on March 30th and 31st 2002. Prior to outplanting many of the *C. laevigatus* individuals appeared weak and spindly. The average maximum shoot height was 23.9 cm prior to outplanting. Surprisingly, by May 15, 2002 the average maximum shoot height for outplanted individuals had dropped to 20.6 cm. However, the plants themselves, on average appear much stouter, healthier and more robust. Only five (1.2%) of, outplanted individuals had died one and a half months after outplanting. The average number of reproductive shoots had also increased from less than 15 to greater than 25 reproductive shoots per outplanted individual.

Due to the smaller stature of the outplanted makaloa seedlings, the low-density plots have not filled in much at this date. It appears that utilizing higher planting densities would be recommended for makaloa.

Cyperus polystachyos

Three hundred individuals of *C. polystachyos* were outplanted on March 17th and 18th, 2002. At the time of outplanting, the average maximum leaf height of *C. polystachyos* seedlings was 48.8 cm. Only one individual contained any reproductive shoots at the time of outplanting. By May 15, 2002 the average maximum leaf height of outplanted individuals had increased to 69.57 cm. As well, outplanted individuals rapidly began flowering and by May 15, 2002 the average number of reproductive shoots per outplanted individual was an astounding 45.8.

Outplanted individuals of this species not only grew rapidly and reproduced rapidly they also had a very high survival rate. To date only 3 individuals of the original 300 have died. Further, this species provides incredibly dense cover; even with only 5 individuals per 0.25 m^2 the canopy cover often approached 90%. In fact it is evident that the high-density plots of *C. polystachyos (*10 plants per 0.25 m^2*)* are much too dense. Individuals in most high-density plots are tall and spindly; in fact plants in some high-density plots are so tall that they are falling over. Plants in the low-density plots appeared much healthier and more similar to *C. polystachyos* individuals occurring naturally.

`Aki `Aki (*Sporobolus virginicus)*

Three hundred and eighty *S. virginicus* seedlings were outplanted on March 22nd and 23rd, 2002. Surprisingly, this species has had the lowest survival rate. By May 15, 2002 thirty-two, or 8.2%, of outplanted seedlings had died. However, many of these dead individuals were located in one block. In fact 25 of the 32 dead individuals were located in block 5. This area appears to be higher in salinity than many of the other blocks in which *S. virginicus* seedlings were planted.

Prior to outplanting, the majority of *S. virginicus* seedlings were quite tall and spindly with very few leaves. In fact they hardly resembled *S. virginicus* plants found growing in the wild. Perhaps this is due to the small (one inch) plugs that they were grown in or perhaps they received too much water at the nursery. The average maximum height of seedlings at the time of outplanting was 25 cm. By May 15th the average maximum height had increased to 31.75 cm. Most notably, or perhaps most importantly, the average number of tillers per individual had increased from 2.8 to 7.3 in the month and a half after outplanting. As well, outplanted individuals appeared much healthier and robust by May 15th, 2002.

`Akulikuli (*Sesuvium portulacastrum)*

Three hundred *S. portulacastrum* seedlings were outplanted on March 22nd and 23rd, 2002. This species has also survived quite well. Only three (1%) of the original 300 plants had died by May 15, 2002. Further, the average number of flowers per plot increased from 0 at the time of outplanting to 7.6 by May 15, 2002. The percent cover per plot increased from an average of 43.15 to 54.75 in this period. This plant appears to be quite hardy and is surviving well in the dry conditions of Pearl Harbor NWR.

Pa`uohi`iaka (*Jacquemontia ovalifolia)*

As mentioned previously, due to nursery error, the 300 seedlings of *J. ovalifolia* were not ready for outplanting until May 12th of 2002. The average height of seedlings at the time of outplanting was 27.4 cm. As of May 31st all of these individuals were still alive and healthy.

VI. Work Remaining

Currently all plant species have high survival rates and plots contain few alien plant species. However, this could rapidly change. In order to survey plant survival and reproduction throughout the year monitoring will continue through March 2003. In addition, plots will continued to be monitored to determine if certain wetland species are better able to compete with or preclude encroachment by invasive plant species. Germination trials (See Appendix II: Research Proposal) will begin in mid-June of 2002 and will continue through February of 2003. At this point all data collected will be analyzed and interpreted. A final report outlining study conclusions and recommendations will be completed and submitted to USFWS.

Appendix 2 – Research Proposal

RESEARCH ON NATIVE PLANTS FOR COASTAL WETLAND RESTORATION ON O'AHU

Agreement 12200-1-J005

To

The U.S Fish and Wildlife Service
Oahu, Hawaii

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I. BACKGROUND

The landscape of the Hawaiian Islands has been drastically altered since the arrival of Polynesian and European settlers. Lowland plains and coastal areas have been particularly susceptible to human alteration. In Hawai'i large areas of natural wetlands and coastal areas have been lost and/or highly modified due to human action (Chang, 1990). In the last century lowland wetland and coastal areas have not only been filled for urban development but they have also been drained or dredged and filled for agricultural purposes such as sugar cane production. Thomas Dahl (1990) estimates that in the 1780's approximately 22,475 acres of wetlands existed within Hawaii's coastal plains. By 1990 the U.S. Fish and Wildlife Service (USFWS) estimated that only 15,474 acres of lowland wetlands remained a loss of 30%. (Dahl 1990, Ducks Unlimited 2000)

Loss of wetland habitat impacts many "ecosystem services" including flood control, groundwater recharge, nutrient cycling, and water purification that are provided by these areas (Mitsch and Gosselink 2000). In Hawaii, one of the most detrimental effects of wetland loss and alteration has been the impact on native waterbird populations. Five of the six endemic Hawaiian waterbirds are currently listed as endangered: the Hawaiian moorhen or 'alae 'ula (*Gallinula chloropus sandwicensis*), the Hawaiian Coot or `alae ke`oke`o (*Fulica americana alai)*, the Hawaiian stilt or Ae`o (*Himantopus mexicanus knudseni),* the Hawaiian duck or koloa maoli (*Anas wyvilliana)*, and the Laysan duck (*Anas laysanensis)* (Shallenberger 1981). All of these species, except the Laysan duck, are found on the main Hawaiian Islands.

In the 1970's the USFWS established several National Wildlife Refuges (NWR) in wetland areas in the Hawaiian Islands. The primary goal of these refuges is to preserve and enhance habitat necessary for protection and recovery of Hawaii's endangered waterbirds. Two of these Refuges, James Campbell NWR and Pearl Harbor NWR are located on the island of O'ahu and encompass approximately 200 acres of wetland habitat (USFWS 2001).

Unfortunately, loss of habitat is not the only threat to the survival of Hawaii's waterbirds. Introduced predators such as the mongoose (*Herpestes auropunctatus)*, cattle egret (*Bubulcus ibis)*, rat (*Rattus* spp.), and feral cat (*Felis* spp.) are a continual threat to waterbird populations. Invasive exotic plants, which degrade wetland habitat, also pose a threat to native waterbirds. Introduced species such as California grass (*Brachiaria mutica*), Indian fleabane (*Pluchea indica*), cattail (*Typha latifolia)*, pickleweed (*Batis maritima)* and California bulrush (*Schoenoplectus californicus*) are aggressive, quick growing species that often form dense monotypic stands that can choke out open water space and cover mudflats; areas necessary for the birds' existence (USFWS 2001). These introduced species also displace native Hawaiian wetland plants and control of these aggressive invasive species is an expensive and time-consuming problem for wetland managers.

Many wetland areas around the state are currently overrun by such exotic and weedy plant species. By 1981 over half (55%) of the species of plants known to occur in Hawaiian wetlands were non-native (Stemmermann 1981 as cited in Chang 1990). Control of these species is necessary to maintain open water and wetland habitat. Recently there has been the desire not only to control invasive species, but also to return native vegetation to wetland sites. Unfortunately, little is known about the life history or ecological requirements of many native wetland plants. Additionally, although wetland and coastal plants have been outplanted in various areas around the state, rarely have they been monitored for survival and reproduction; records of success or failure are largely anecdotal.

This research project will identify native plants that may be useful for wetland restoration, evaluate outplanting techniques that may aid survival of outplanted species, and determine environmental conditions that support germination, growth, and reproduction of these native plants. Results of these studies will not only help identify native plants that may be useful for wetland "restoration" due to their ease of propagation and ability to survive outplanting, but will also provide information that will aid in establishment and recruitment of these native species.

II. OBJECTIVES

1. Summarize literature, including paleobotanical studies, and ongoing wetland restoration projects in Hawaii that focus on outplanting and/or optimizing recruitment of native plant species.

2. Identify native plants that are able to survive and reproduce over time with a minimum of human intervention

Native wetland species, sedges and groundcovers, will be outplanted and monitored for survival and reproduction to determine which species have the highest survival, establishment and reproductive rates.

3. Identify native plants that are able to resist and/or effectively out-compete invasive weeds.

Prior to outplanting, all vegetation will be removed from each plot, including control plots. Subsequently, outplanted plots of native species will undergo either a "competition" or "no competition" treatment. The "competition" treatment, in which non-planted species are not removed after the initial plot clearing, will help determine which species are best able to effectively compete with other plants, including invasive weeds. Non-planted species in the "no competition" plots will be removed monthly. This will help determine if weeding ("no competition") is necessary for establishment of outplanted native wetland plants. In addition, this treatment will test the hypothesis that plants not subjected to competition will demonstrate higher growth and survival rates than plants subjected to competition with non-planted species.

4. Determine whether initial planting densities affects establishment of native wetland plant species and their ability to compete with alien species.

Native species will be planted in different planting densities to test the hypothesis that higher initial planting density will reduce invasion and establishment of alien species.

5. Assess the reproductive phenology of outplanted species.

Plants will be monitored to determine whether vegetative growth, flowering and seed production is correlated with environmental conditions of temperature, rainfall and soil moisture. This information will help determine whether the plant species under investigation demonstrate seasonality in their reproduction.

6. Determine the relationship between soil moisture and salinity on seed germination response of wetland sedges.

Germination trials under different soil moisture regimes will test the hypothesis that percent germination of wetland sedges will be highest under moist soil conditions and will decrease with increasing standing water depth. It is also hypothesized that germination will be higher under moist soil conditions than dry soil conditions. Germination trials under different salinity regimes will test the hypothesis that germination percentage will decrease under increasing salinity.

7. Provide native plant restoration recommendations to natural resource managers.

8. Assess, through literature, observation and communication with wetland practitioners, the potential for native plants to provide food, shelter and nesting material for endangered waterbirds.

III. JUSTIFICATION

Little is known about restoring native plants to wetland habitats in Hawaii. Restoration of native vegetation to Hawaiian wetlands will not only return the area to a more "natural" state, but also could improve native waterbird habitat and reduce the need for invasive plant control efforts. This project will add to the scant information regarding native plants that may be useful to wetland restoration efforts, outplanting techniques that will increase survival of outplanted species, and environmental parameters that affect growth, survival and reproduction of native wetland plants.

Outplanting of native species is potentially one of the quickest ways to restore native vegetation to an area. Unfortunately, transplanted individuals do not always survive, especially without continual maintenance. Further, aggressive alien weeds may invade a restoration site and replace native vegetation. Experimental outplanting trials will help assess which native wetland species are best able to survive, grow and reproduce with minimum maintenance. Additionally, utilizing various planting densities may help determine if initial planting density can help limit re-colonization by invasive weeds. Presumably, densely planting native species will help minimize encroachment by alien species. Experimentally testing this hypothesis will help establish whether, in the long run, outplanting native species in dense stands is worth the initial cost.

Outplanted species will also be monitored for reproductive status. In order to restore native vegetation to an area it is useful to understand the phenological traits of individual plant species.

Plant vegetative growth, flowering and reproductive output shape plant population dynamics. Monitoring the temporal occurrence of phenological events such as vegetative growth, flowering and seed production is important for assessing environmental conditions which may influence these events. Phenological information can provide land managers with vital information regarding timing of vegetative production and seed production that can help guide management decisions, such as when to conduct vegetation control and how abiotic conditions impact production of native wetland plant species.

Understanding specific germination requirements of native and alien species is also important for any restoration effort. Sedges and other wetland species often spread primarily by vegetative or clonal growth (van der Valk et al. 1999). However, initial establishment of a new population or colonization of a nearby area is through seed dispersal and germination. Seeds of many wetland species germinate in response to changes in hydrology such as continued drawdown or flooding (LaDeau and Ellison, 1999). In fact many emergent wetland species require bare mudflats or shallow water for germination and seedling establishment (ter Heerdt and Drost 1993). Salinity is another factor that may influence germination of emergent wetland species (Martinez et al. 1992). Seeds of many wetland species are unable to germinate under conditions of high salinity, high water and/or low light. Determining the influence of abiotic conditions, such as soil moisture, water level, and salinity on germination rates is vital not only for restoration efforts but also for those attempting to manage for native plant communities.

Understanding native bird use of native wetland plant species will help determine native wetland plant species useful to Hawaii's four endangered waterbirds. Although several species of alien plants are currently being utilized as food, cover and nesting material by native waterbirds, these aggressive plant species form dense stands that choke out open waterways and require continual maintenance and control. Native wetland species may prove to be less invasive and just as attractive to waterbirds as these invasive species. This project will begin to assess which native wetland plant species could potentially be utilized by native waterbirds as sources of food, cover and nesting material.

IV. PROCEDURES:

A. Permits:

A Special Use Permit will be issued through the Oahu National Wildlife Refuge Complex.

B. Methods:

1. Study Area:

Work will be conducted at the Honouliuli Unit of Pearl Harbor National Wildlife Refuge (NWR) (See Appendix I). The Honouliuli Unit of Pearl Harbor NWR consists of two brackish water ponds. Outplanting will occur on the margins of Pond 2 at Honouliuli.

Study blocks will be marked with small (40 cm) blue flags. Approximately 2000 plants will be marked with numbered aluminum tags that will be tied to the base of each plant. All markers and tags will be removed upon completion of the study (ca.one year).

2. Techniques:

a. Plant Background

Seven species of native plants will be utilized during this project. All species are indigenous to Hawaii and occur in wetland areas. Four sedge species: `ahu `awa (*Cyperus (Cyperus) javanicus*), makaloa (*Cyperus laevigatus*), makai (*Bolboschoenus maritimus*), and *Cyperus (Cyperus) polystachyos* will be used in the shallow water and mudflat regions of the pond edge. These sedges have the capacity to serve in soil stabilization, provide coverage that will hopefully reduce colonization by invasive weeds and provide food, cover and/or nesting material for waterfowl. One grass, `aki`aki (*Sporobolus virginicus),* and two perennial groundcovers: `akulikuli (*Sesuvium portulacastrum)* and pa`uohi`iaka (*Jacquemontia ovalifolia*) will be planted higher up on the banks above the waterline. These groundcovers will also be useful in soil stabilization and reduction of colonization by invasive weeds.

`Ahu`awa (*Cyperus (Cyperus) javanicus*) currently grows at Pearl Harbor NWR although it is not dominant (pers. obs.). It is apparently a re-colonization within the refuge ponds and has only recently been observed (Silbernagle, pers. comm. 2001). It is often found in coastal marshes exposed to salt or brackish water as well as freshwater marshes and taro paddies (Stemmermann 1981). Previous studies have found `ahu`awa to be a fast growing and hardy plant that can tolerate drought as well as standing water (Koob 1999).

Makaloa (*Cyperus laevigatus)* is not known from Pearl Harbor NWR, but is found in other wetlands around the state (Silbernagle pers. comm.; pers. obs.). This sedge may provide a food source to native waterbirds. Makaloa occurs on mudflats, sandy coastal sites and on edges of fresh, brackish and saltwater ponds (Stemmermann 1981). This sedge survives equally well in shade house or full sun (Koob 1999). Makaloa may grow faster when grown in standing water or kept very wet (Koob 1999).

Makai (*Bolboschoenus maritimus),* a large perennial sedge, is tolerant of saline conditions. This species rapidly reproduces and can produce as many as 45 tillers from a single corm (Kantrud 1996). Further, coots and moorhens have been noted to nest in stands of makai (Silbernagle, pers. comm. 2001). Seeds of this sedge may also provide a food source for waterbirds. Makai currently grows quite abundantly in the Honouliuli Unit of Pearl Harbor NWR.

Cyperus (Cyperus) polystachyos is a perennial sedge, which is currently represented by just a handful of plants at the Honouliuli Unit of Pearl Harbor NWR. Coots have been observed feeding on seed heads and young leaves of these plants at James Campbell NWR (Silbernagle, pers. comm. 2001). According to Stemmermann (1981), *C.*

polystachyos is found in cultivated wetlands and ruderal wetlands as well as areas under occasional influence of brackish water.

`Aki`aki (*Sporobolus virginicus*) is a native perennial grass with buried rhizomes and erect branches to 30 cm tall (Stemmermann 1981). It may be attractive to waterbirds as a food source, and if grown in dense stands, may help shade out invasive species. `Aki `aki is found both in coastal marshes as well as along streambeds. This species may survive equally well when kept constantly moist or allowed to dry out between watering (Koob 1999). ‘Aki`aki has been seen growing at Pearl Harbor NWR, but is not abundant.

‘Akulikuli (*Sesuvium portulacastrum*) is a perennial, succulent herb common in wetland areas. It is saline tolerant and often grows in coastal wetlands and mudflats. It is a facultative wetland species found in varying coastal habits including edges of fresh, brackish and saltwater ponds (Koob 1999). It generally occupies habitat above the waterline, but is tolerant of periodic inundation. As a hardy groundcover, it will be used to vegetate embankment slopes for soil stabilization and to shade out invasive species.

Pa`uohi`iaka (*Jacquemontia ovalifolia)* is another perennial groundcover currently found in wetland and coastal areas of Oahu. It is a sprawling vine that is tolerant of salt and mild drought (White 1999). The plant is fast growing and requires minimum water and maintenance and its dense cover may be useful in shading out emergent invasive species (White 1999).

b. Outplanting Field Trials:

Seedlings of seven native wetland plant species will be outplanted in single species blocks. A randomized block design will be utilized with ten 1.25 m x 6.5 m blocks for each of four sedge species and ten 2 m x 3.5 m blocks for each of three groundcover species. This equates to a total of 40 1.75 x 6.5 m blocks for sedge species and 30 2 x 3.5 m blocks for groundcover species. Each block will contain 6 permanent 0.25 m^2 plots, for a total of 420 permanent 0.25 m^2 plots (See Attachment 1). The overall experimental design will be a 2 x 2 factorial, with treatments of high density / low density and competition / no competition. Thus, each permanent plot will be planted in one of six different arrangements [high density/no competition, high density/competition, low density/no competition, low density/competition and control I (no species planted, no competition) and control II (no species planted, competition)]. Each block will contain one replicate of each treatment for each species. Thus, there will be a total of ten replicates of each combination of planting density and competition for each species. Plants in each 0.25 m^2 plot will be monitored over the course of the project (ca. 1 year) to evaluate the survival, establishment, recruitment and reproductive phenology of each species.

Based on the size of the seedlings, species will initially be planted at three different densities:

1) *Cyperus (Cyperus) polystachyos, Cyperus laevigatus, Jacquemontia ovalifolia,* and *Sesuvium portulacastrum* will be planted at low density = 5 plants/0.5 m^2 and high density = 10 plants/0.5 m^2
2) *Bolboschoenus maritimus* and *Cyperus javanicus* will be planted at low density = 4 plants/0.5 m^2 and high density = 9 plants/0.5 m^2
3) *Sporobulus virginicus* will be planted at low density = 6 plants/0.5 m^2 and high density = 13 plants/0.5m^2
4) In the control plots, all vegetation will be removed as in the other plots; however, nothing will be planted in these plots.

Blocks will be chosen from sites around the edge of Pond 2 at Pearl Harbor NWR. Prior to any manipulation, baseline data consisting of current vegetation and soil characteristics (salinity, soil moisture and soil temperature) will be taken. Next, blocks will be manually cleared of all alien species. Once a month, non-planted species will be removed from all "no-competition" plots. Preparation of sites and outplanting of native species will take approximately two weeks, beginning on March 8 and concluding on approximately March 23, 2002.

i. Native Species Monitoring

Twice a month, for one year after outplanting, plots will be monitored to assess survival and growth of native species. Monitoring will be conducted over a four-day period twice a month. Each outplanted individual / ramet will be checked for survival, vigor and reproductive/vegetative status. Depending on growth form and pattern, specific measurements for growth and reproduction will vary for different species.

Growth:
Recruitment and growth of `aki `aki (*Sporobolus virginicus),* makai (*Bolboschoenus maritimus)*, and `ahu `awa (*Cyperus javanicus)* will be measured by counting the number of shoots (tillers) produced and the height of the longest shoot or leaf of each ramet. Percent cover will also be recorded so that change in percent cover over time can be documented. Makaloa (*Cyperus laevigatus)* and *Cyperus polystachyos* produce too many shoots (tillers) to accurately count, thus, growth for these species will be monitored through percent cover and measurement of the longest stem / leaf of each tagged ramet. 'Akulikuli (*Sesuvium portulacastrum)* and pa`uohi`iaka (*Jacquemontia ovalifolia*) both exhibit sprawling growth with stems often rooting at the nodes, making it difficult to tell individuals apart. Thus, growth for these two species will be assessed using percent cover only.

Reproduction:
The number of reproductive stalks will be recorded for `aki `aki (*Sporobolus virginicus),* makai (*Bolboschoenus maritimus)*, *C. polystachyos* and `ahu `awa (*Cyperus javanicus)* during each monitoring period. Because makaloa (*Cyperus laevigatus)*, produces large numbers of reproductive shoots, the number of reproductive shoots will be estimated. Number of flowers per plot will be recorded for 'akulikuli (*Sesuvium portulacastrum)* and pa`uohi`iaka (*Jacquemontia ovalifolia*) will be charted by counting the number of flowers per plot. Reproductive stage will be documented.

Abiotic factors:
Water depth, salinity, soil temperature, soil moisture and rainfall will also be measured in each plot twice a month. To promote establishment of outplanted seedlings, each plot will be given one gallon of supplemental water per week for the first month after outplanting. After the first month, plots will be given one gallon of water every two weeks for the next two months.

ii. Non-planted and non-native species monitoring*:*

Plots will be monitored for appearance, establishment and species composition of species not planted in the plots. Species composition and percent cover for any non-planted species will be recorded for each plot. After percent cover for each non-planted species is documented, these species will be removed from the "no-competition" treatments once a month by clipping plants at ground level. Plant clippings will be placed in separate paper bags by species and allowed to dry. Dry biomass weights will be measured and recorded to assess differences in biomass of non-planted species in low vs. high-density and control treatments.

c. Germination trials

Seeds from native wetland sedges will be collected periodically throughout the year and will be tested for germination rates under different conditions. Seeds of four sedge species, *Cyperus javanicus, Cyperus polystachyos, Bolboschoenus maritimus,* and *Cyperus laevigatus* will be utilized in the germination trials. Seeds will be collected from outplanted plants as well as natural populations at the Honouliuli Unit of Pearl Harbor NWR. Seeds will be placed under experimental conditions in growth chambers at the University of Hawai'i at Manoa within one week of collection. Germination trials will begin in mid-June 2002 and will be run every three months thereafter through March 2001.

Each combination of species, soil moisture and salinity levels as described below will be carried out with five replicates of twenty seeds. After three weeks (following Baskin and Baskin 1998), ungerminated seeds will be tested for viability using the tetrazolium chloride test (Baskin and Baskin 1998). In this test, embryos are either dissected from the seed or seeds are cut so that the embryo is bisected. Embryos are then placed in a 0.1% solution of 2,3,5-triphenyl-2H tetrazolium chloride (TTC). In the presence of TTC, a viable embryo turns red or pink (Baskin and Baskin 1998).

During all germination trials, seeds will be kept in growth chambers with alternating 12 h light / 12 h dark. Temperatures will alternate between approximately 30 degrees Celsius and 15 degrees Celsius corresponding with the light/dark regime. Thermometers will be placed out in the field prior to germination trials to estimate the minimum and maximum temperatures occurring in the field at the time of seed collection.

i. Soil Moisture:

Germination response of wetland sedges under different moisture levels will also be assessed. Four water levels: 1 cm of standing water, saturated, moist but not saturated and dry will be tested. Seeds will be placed on blotter paper in petri dishes under the above conditions. Five replicates of twenty seeds for each species and treatment will be tested. Water will be added to dishes daily to maintain desired soil moisture level. Germinated seeds will also be counted and removed daily.

ii. Salinity:

Germination trials under 4 different levels of salinity (0.0, 0.5, 1.0, 2.0% NaCl) will also be conducted to determine the ability of native wetland species to germinate under levels of salinity that might be found at Honouliuli and in other brackish wetlands. Twenty seeds of each species will be placed in a Petri dish on blotter paper saturated with distilled water or with the appropriate saline solution. Petri dishes will be tightly sealed to reduce evaporation of water, which could possibly alter the conditions of salinity. Water levels will be checked daily to ensure that evaporation is not occurring. Germinated seeds will be counted daily and then removed from the petri dish. After 3 weeks un-germinated seeds will be tested for viability.

d. Waterbird use of native plants

Because the outplanted plots of native species are relatively small, it is unlikely that native birds will visit these stands heavily. Thus, personal communication with wetland managers and researchers and literature reviews will supplement observational study of native plant use by native waterbirds. Observational study will include a five minute scan/observation period after every hour of plant monitoring / field work. Bird behavior, such as feeding, resting or nest building as well as vegetation type in which behavior is occurring will be noted. Further, Mike Silbernagle (USFWS Biologist) and his volunteers will be given a brief form to fill out during their monthly bird counts and surveys. This form will chart sightings of native birds in stands of native vegetation, duration in these stands and evidence of feeding, resting in or using native plants for nesting material.

3. Analysis

a. Outplanting field trials

Native Species Monitoring

Depending on species, percent survival, average percent cover, average number of shoots produced, average length of longest shoot/leaf and average number of reproductive shoots per individual/plot will be calculated at the end of the year.

A multi-way ANOVA (analysis of variance) model will be used to test the effect of planting density and competition on survival (percentage survival per plot), shoot production (average of total stem production per plant), reproduction (average number of reproductive stems/flowers per plant), average shoot height and change in percent cover of native species (initial percent cover – final percent cover). All tests of significance will be conducted at the $p < 0.05$ level. Multiple regression analysis will be used to determine whether abiotic conditions of water depth, soil moisture, soil salinity, soil temperature and rainfall are correlated with survival, shoot production, reproductive rates and density of outplanted individuals.

To determine seasonality of reproduction 95% confidence intervals will be computed. For makai *(Bolboschoenus maritimus)*, \`aki \`aki *(Sporobolus virginicus), Cyperus polystachyos* and \`ahu \`awa *(Cyperus javanicus)* confidence intervals will be computed for the proportion of shoots in vegetative versus reproductive stages on monitoring dates throughout the study period. For \`akulikuli (*Sesuvium portulacastrum)*, pa\`uohi\`iaka (*Jacquemontia ovalifolia)* and \`aki \`aki (*Sporobolus virginicus)*, the average number of flowers per plot for each monitoring period will be calculated.

Non-native Species Monitoring

A one-way ANOVA model will be used to assess whether final percent cover of non-planted species (in the competition plots) is significantly different ($p < 0.05$) in the high-density, low-density and control treatments. This test will help assess whether initial planting density is correlated with a difference in appearance and establishment of non-planted species.

A one-way ANOVA will also be used to assess whether biomass of non-planted species is significantly different ($p < 0.05$) in high-density, low-density and control treatments. Biomass will be broken down by individual species as well as overall biomass, since certain species weigh more than others.

b. Germination data

Germination data will be analyzed separately for each species and each treatment using a one-way ANOVA (analysis of variance) to test for any effect of salinity and soil moisture on germination percentages. Germination data will be arc-sine transformed prior to running ANOVA tests. All tests of significance will be conducted at the $p<0.05$ level.

c. Waterbird Use of Native Plants:

Observations, data forms, literature and personal communications will be summarized. Time spent in each stand of native plant as well as uses of native plants will be determined and any differences between plant species will be noted.

4. Interpretation:

A. Outplanting data:

Differences in survival rates of the native wetland plant species utilized in the outplanting trials will provide knowledge to guide further restoration projects. Species that have low survival and reproductive rates might not be considered for future projects, or might require further investigation into methods for improving survival and production. Determining whether initial planting density results in differences in survival rates, production, final density and reduced establishment of non-native species will help decide whether, in the long run, restoration efforts will be enhanced by initially planting dense stands of native species. Assessing whether competition from non-planted species reduces survival and growth of planted species will help managers determine whether periodic weeding is necessary to promote the establishment of outplanted species. Finally, correlations between survival, growth and density of native species under varying environmental conditions of salinity, soil moisture, and temperature will help guide decisions on suitable geographic areas for further restoration efforts.

B. Germination data:

Knowledge of differences in percent germination under different conditions of soil moisture and salinity can assist land managers in determining levels of soil moisture and salinity under which native plants can and cannot germinate. Once again this can help inform management decisions such as how timing and duration of water drawdown may encourage/discourage germination of native species. It will also help inform further restoration efforts that may include the use of seed to establish populations of native species.

C. Waterbird Use:

Information collected regarding waterbird use of native vegetation will also help shape wetland management efforts. Plants deemed desirable to endangered waterbirds, due to their use as a food source, nesting material and shelter, may be chosen or planted in higher abundance than those deemed less desirable or useful.

V. PERSONNEL

Karen Brimacombe – Project Leader
University of Hawai'i at Manoa

Mike Leech – Outplanting assistant
DLNR – Division of Forestry and Wildlife

Mike Silbernagle – Periodic help with bird monitoring
USFWS

VI. COSTS

No additional costs to the U.S. Fish and Wildlife Service should be incurred during the course of this study.

VII. SCHEDULE

This project began on August 1, 2001. Fieldwork and bird monitoring began March 9, 2002 and will end approximately one year from this date. Germination trials will begin in June of 2002 and will take place approximately every three months thereafter until March of 2003.

VIII. REPORTS

Quarterly progress reports will be completed 3, 6, and 9 months after the project begins. A "year-end" report will be due 12 months after the project begins. Within one year of the project completion date a copy of the completed Master's Thesis submitted to the University of Hawai'i at Manoa will also be submitted to both USFWS-Ecological Services and Oahu National Wildlife Refuge Complex Headquarters.

IX. PUBLICATIONS

If publishable data is obtained at the end of the project an article will be submitted to a peer-reviewed, scientific journal. At least one complimentary copy of any publications accepted by a scientific journal will be sent to the Oahu NWR Complex Headquarters.

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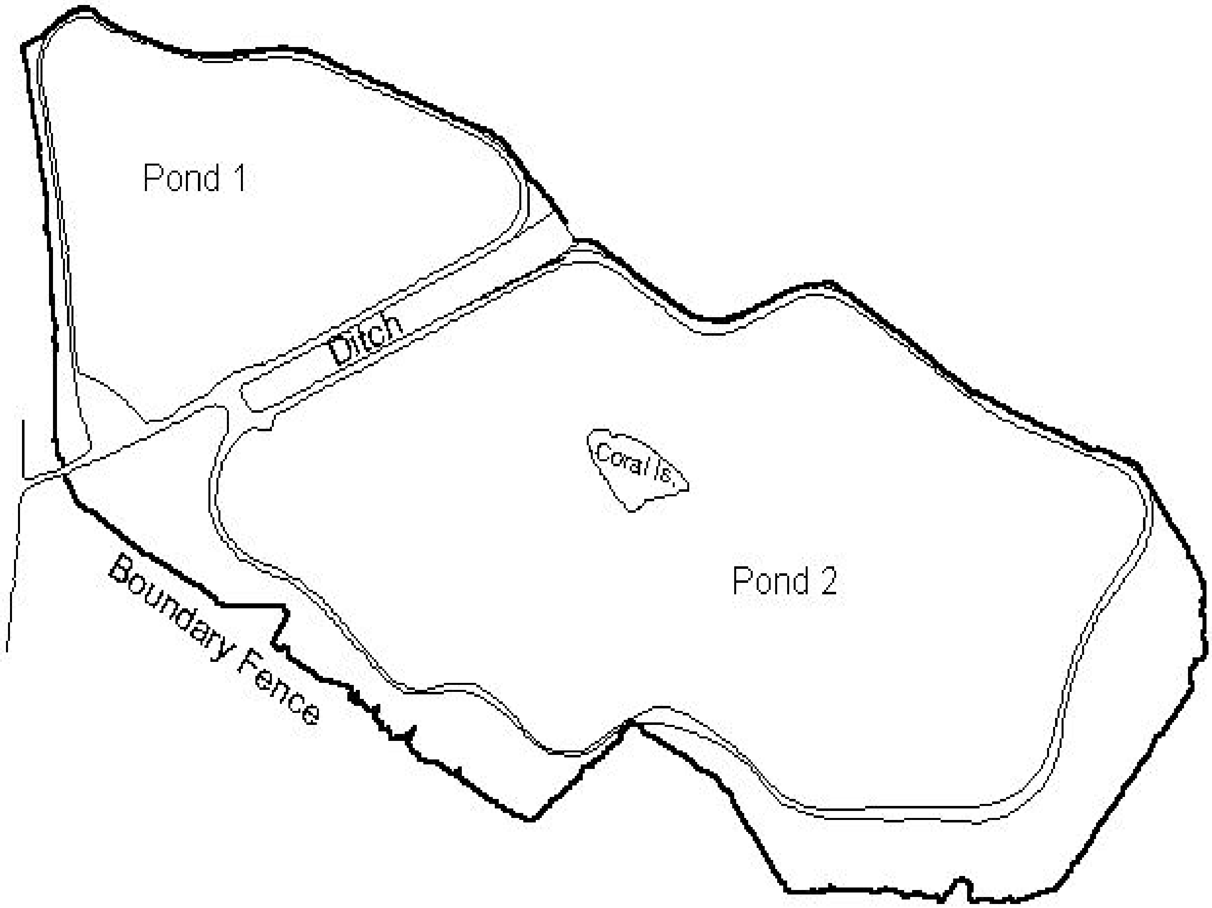
Appendix III – Map of Honouliuli Unit, Pearl Harbor NWR

Pond Outline – Honouliuli Unit, Pearl Harbor NWR

Outplanting plots are located around the perimeter of pond 2.


Pond 1
Ditch
Coral Is.
Pond 2
Boundary Fence

Appendix III – Wetland Restoration in Hawaii (include??)

UNIVERSITY OF HAWAII AT MANOA
Office of Research Services
Sakamaki Hall 200, 2530 Dole Street,
Honolulu HI 96822

Pacific Cooperative Studies Unit
University of Hawaii at Manoa

RESEARCH ON NATIVE PLANTS FOR COASTAL WETLAND RESTORATION ON O'AHU

Quarterly Report for First Three Months

Agreement 12200-1-J005

To

The U.S Fish and Wildlife Service
Oahu, Hawaii

Karen Brimacombe
Project Researcher
Department of Botany
University of Hawaii

Introduction

Wetland ecosystems provide extremely important "services" such as flood control, water quality improvement, groundwater recharge, minimizing erosion and decreasing sedimentation of coastal waters (Mitsch and Gosselink 2000). Unique recreational and educational opportunities are also provided by these ecosystems, and in Hawai'i, wetland areas are vital to the preservation of four endangered waterbirds. Until the mid 1970's, however, destruction of wetlands was a common and accepted practice in the United States (Mitsch and Gosselink 2000). Wetlands were drained, dredged, ditched, and filled for development of agricultural fields and urban development. Hawai'i did not escape these practices and by 1990 approximately 31% of Hawaii's coastal wetlands had been lost or altered for agriculture and development (Dahl 1990).

Most of Hawaii's remaining coastal wetland habitat is degraded due to invasion by non-native plant species. Fast-growing invasive species, such as California grass (*Brachiaria mutica),* Indian fleabane (*Pluchea indica*), pickleweed (*Batis maritima)*, and California bulrush (*Scirpus californicus*), often encroach upon open water and form monocultures too dense for waterbirds to penetrate. Not only do alien species impair habitat for native and migratory waterbirds, they also pose a dire threat to native wetland plant populations and can alter ecosystem structure. Removal and control of alien plant species is an expensive and time-consuming process. "Restoration" efforts in many existing wetlands have focused on control and removal of non-native species. Recently, however, there has been a push to include reintroduction of native vegetation to wetland restoration efforts. Unfortunately, rarely have the successes (and failures) of wetland restoration in Hawai'i been documented.

One difficulty in planning and implementing any “restoration” project is the struggle to determine the previous or “natural” condition of a heavily altered ecosystem. Further, as all communities and ecosystems are continually changing, there is the difficulty of discerning how far back in time to go when choosing species to re-introduce. Surveying pristine wetland areas or perusing historic accounts may offer information about the “natural” state of these areas. Unfortunately, there are few pristine wetland areas left in the state and historically, many lowland wetlands were altered with the arrival of Polynesian settlers. However, recent paleobotanical work has provided interesting and important insight into pre-human vegetation communities in lowland areas of the Hawaiian Islands. Information gleaned from these studies may be useful for development of restoration plans.

Paleobotanical Studies

Lowland areas on all main Hawaiian Islands have been extensively altered and modified by both Polynesian and European settlers. In fact Athens (1997) conjectured that humans had extensively altered almost 100% of the landscape below 1, 5000 feet by the 1600’s. Following Cuddihy and Stone (1990), Athens (1997) states: “the vegetation in the few remaining “wild” lowland areas of O’ahu is absolutely different from what would have been in these areas in pre-human times”. Paleobotanical studies can provide insight on these pre-human plant communities. As well, these studies help document vegetation changes initiated by Polynesian and European settlers. Palynological work by Athens, Ward, Burney and others has provided much information on prehistoric vegetation of lowland areas of the Hawaiian Islands.

Paleobotanical Evidence: O’ahu

In the early 1990’s Athens and Ward took wetland sediment cores from 12 locations on O’ahu including Kawainui Marsh, Hamakua Marsh, Ft. Shafter flats, ‘Uko’a Pond and Pearl

Harbor (Athens and Ward 1993, 1997). These cores provide a continuous lowland sedimentary record that dates from well before initial Polynesian occupation until well after colonization, with some cores including post-European colonization. Of these 12 samples, cores from Kawainui and Ft. Shafter have been the most completely analyzed for pollen and plant macrofossils (Athens et al. 1992).

Over 88 species or types of angiosperm pollen were found in samples from Kawainui Marsh and 84 species were found from Ft. Shafter Flats (Athens and Ward 1993). Approximately 65-70 percent of pollen types at Ft. Shafter and Kawainui were of Dry-Mesic Forest species, 28-29% were Mesic-Wet Forest species, and only 2% were grasses and sedges from the coastal marsh community (Athens et. al. 1992). The pollen and fossil remains analyzed by Athens, Ward and Wickler (1992, 1997) indicate that leeward forests were dominated by an upper story of *Pritchardia spp.* with a diverse secondary and understory of *Acacia koa, Antidesma spp., Chenopodium oahuense, Dodonaea viscosa, Kanaloa spp., Erythrina sandwicensis, Colubrina spp. and Gouania spp.* Although similar species were found at the windward site, lowland windward cores were characterized by three dominant pollen types: *Pritchardia, Dodonaea viscosa,* and *Kanaloa spp.* The abundance of tree and shrub pollen uncovered in the sediment cores indicates that a forested ecosystem dominated lowland areas of O'ahu in pre-human times (Athens 1997).

The abundance of Loulu palm (*Pritchardia* spp.) pollen found in early (~1200 B.C. to ~1200 A.D.) sediment layers is surprising. Currently, *Pritchardia* palms on O'ahu are restricted to isolated valleys and gulches. Based on pollen evidence, Athens (1997) believes *Pritchardia* spp. was a dominant taxon in the lowlands "virtually everywhere on O'ahu" prior to human arrival. The abundance of *Kanaloa kahoolawensis* pollen in both leeward and windward sites is also interesting as currently only two plants of *K. kahoolawensis* are known to exist in all of Hawai'i (Lorence and Wood, 1994).

Abundance of *K. kahoolawensis* and *Pritchardia spp.* pollen began to decline sharply between 1000 and 1200 A.D (Athens 1992). Concurrently, pollen frequency of native and introduced weedy herbs and sub shrubs of Chenopodium and Amaranth families and grasses of the Poaceae family began to rise dramatically (Athens 1992). This change is indicative of disturbance, possibly indicating clearing of land for agricultural purposes, introduced plant diseases, and/or the introduction of the Polynesian rat (*Rattus exulans),* which is believed to have predated on the seeds of *Pritchardia* spp. (Athens 1997). At Kawainui Marsh the abundance of sedge pollen also increased sharply after about 1000 A.D. (Athens 1992). This rise in sedge pollen may indicate deforestation in areas surrounding Kawainui Marsh (Allen 1997).

Paleobotanical evidence: Kaua'i

Paleoecological work by Burney et al. (2001) on the south coast of Kaua'i also revealed an abundance of *Pritchardia* spp. pollen in pre-human sediment. Burney et al. (2001) took cores in a large sinkhole that had been a fresh to oligohaline lake or marsh during the mid-Holocene. Pollen remains indicate that plants such as naupaka (*Scaevola sericea)*, pa`uohi`iaka (*Jacquemontia ovalifolia*) and hala (*Pandanus tectorius)* that currently exist in coastal and strand vegetation communities were present in the mid-Holocene. Further, although not identified to genus or species, there was a high frequency of Poaceae and Cyperaceae pollen (Burney et al. 2001).

However, plants currently considered xeric, mesic or wet forest species also appear to have coexisted in lowland areas in the mid-Holocene (Burney et al. 2001). Pollen and seeds of species currently found in low elevation dry and mesic habitats, such as 'akia (*Wikstroemia uva-ursi*), 'a'ali'i (*Dodonea viscosa)*, soap berry (*Sapindus oahuense)*, hao (*Rauvolfia sandwicensis*), and sandlewood (*Santalum freycinetianum*) were uncovered from mid-Holocene sediment layers at the Kauai site (Burney et al. 2001). Macrofossils of plant such as *Kokia kauaiensis, Ochrosia kauaiensis, Pteralyxia kauaiensis* and *Zanthoxylum* spp. that are currently rare and/or restricted to high elevations were also present (Burney et al. 2001).

As expected, sediment layers from the period of Polynesian settlement show remains of coconut (*Cocos nucifera)*, bitter yam (*Dioscorea bulbifera),* olona (*Touchardia latifolia)*, and other plants brought to Hawai'i with Polynesian settlers. Sediment layers dated to post-European contact contained an abundance of koa haole (*Leucaena leucocephala),* kiawe (*Prosopis palida)* and Java plum (*Syzygium cumini)* seeds (Burney et al. 2001). These species, all European introductions, are now widespread in lowland areas of Hawai'i.

Discussion

Paleoecological studies are useful in providing insight into the vegetative history of an area however these studies have limitations. First, wind-pollinated species, such as *Pritchardia* spp., that produce large amounts of pollen may be over represented in the pollen assemblage (Jackson 1997). Second, remains of sedges and grasses, which offer the most insight into the history of wetland vegetation, often cannot be identified to genus, species or even the family level (Jackson 1997). Lastly, pollen in sediment layers may collect from the entire watershed making it difficult to pin down the exact flora of a small-scale site such as a wetland proper. Despite these limitations paleobotanical studies offer interesting and useful information. One of the most interesting conclusions drawn from these studies is the notion that a forested ecosystem dominated the lowland areas throughout the Hawaiian Islands. Given this scenario, prior to human colonization many lowland wetlands were probably swamp-like ecosystems with a tree and shrub canopy and fern, grass and sedge understory. This is vastly different than the open coastal plains and wetlands of the present day.

Wetland Restoration in Hawai'i

Wetland "restoration" projects in Hawai'i include wetland mitigation projects, as well as coastal and upland habitat improvement projects. The goal of many wetland "restoration" projects is to establish or enhance habitat for Hawaii's endangered waterbirds. Thus, creation of new ponds and waterways and/or removal of non-native species have been the primary focus of many "restoration" efforts. Very few projects in the state have attempted reintroduction of native plant species.

Although many state, federal and non-profit agencies have begun to implement wetland "restoration" projects in Hawai'i, rarely has information regarding design and results of these efforts been printed or published. Gathering information concerning current and past wetland projects usually requires personal communication with individuals involved in these efforts. The following review provides an island-by-island guide to the various wetland "restoration" efforts in Hawai'i.

Oahu

Kawainui Marsh

Encompassing approximately 900-acres, Kawainui Marsh is the largest remaining freshwater wetland in the state of Hawai'i (Elliott and Hall 1977). Two community based groups, 'Ahahui Malama i ka Lokahi and Kawai Nui Heritage Foundation, have been working on "restoring" a 12-acre area of the marsh, known as Na Pohaku o Hauwahine, since 1997 (http://www.aecos.com/aml/KBAC.html 6 Jan. 2002). Besides removal of alien vegetation, which has helped open up new waterways, several native plant species have been 2planted at this site. Native species outplanted include: 'ae 'ae (*Bacopa monnieri),* neke (*Cyclosorus* *interruptus),* kaluha/makai (*Bulboschoenus maritimus)*, and makaloa (*Cyperus laevigatus)* (http://www.aloha.net/~cburrow/KNHF_7.html, 12 Dec. 2001).

In January of 2000 a single planting of makaloa (*Cyperus laevigatus)* was attempted and although the planting had spread and set seed by April, continued growth did not occur and seedlings have failed to thrive (http://www.aloha.net/~cburrow/KNHF_7.html, 12 Dec. 2001). Although abundant in the marsh, the indigenous fern neke (*Cyclosorus interruptus)* also did not respond well to transplanting efforts. On the other hand, plantings of both the ‘ae ‘ae (*Bacopa monnieri)* and kaluha/makai (*Bulboschoenus maritimus)* were quite successful. In May 2000 mats of ‘ae ‘ae were transplanted from one area of Kawainui to the restoration site (http://www.aloha.net/~cburrow/KNHF_7.html, 12 Dec. 2001). These ‘ae ‘ae mats rapidly spread and now cover a large area of the cleared marsh border and pond margins. The dense growth of ‘ae ‘ae (*Bacopa monnieri)* has been able to exclude seeding by weedy species. Three small plants of kaluha/makai (*Bulboschoenus maritimus),* taken from Kapiolani Park and transplanted in June of 2000 have rapidly spread by both seeds and runners (http://www.aloha.net/~cburrow/KNHF_7.html, 12 Dec. 2001).

Other native species that have been outplanted include: ‘akia (*Wikstroemia uva-ursi),* akoko (*Chamaesyce celestroides),* ewa hinahina (*Achyranthes splendens)*, kulu’i (*Nototrichium sandwicense),* naio (*C.yoporum sandwicense)*, ‘ohelo kai (*Lycium sandwicense),* pa’uohi’iaka (*Jacquemontia ovalifolia),* and pohinahina (*Vitex rotundifolia),* (http://www.aecos.com/aml/PlantListing.html, 25 October 2001; Eric Guinther, personal communication, January 2001). The pa’uohi’iaka, pohinahina, ewa hinahina, ‘ohelo kai, and kulu’i grew well without supplemental water, once established. However the ‘akia, ‘akoko, and naio did not fair as well and required supplemental water and tending in order to survive (Eric

Guinther, personal communication, January 2001; http://www.aecos.com/aml/PlantListing.html, 25 October 2001).

Recently, ‘Ahahui Malama i ka Lokahi has received a grant from the Kailua Bay Advisory Council to study and carry out restoration efforts in Kawai Nui Marsh (http://www.aecos.com/aml/KBAC.html 6 Jan 2002). Plans for the grant include characterizing the physical and chemical components of the marsh, sampling the biota to determine distribution of aquatic species, and to restoring water bird habitat on an islet near the Oneawa outlet of the marsh. The end goal is to provide an education/demonstration site that incorporates native vegetation, nature trails, and varied habitat for wetland flora and fauna (http://www.aecos.com/aml/KBAC.html 6 Jan 2002).

Hamakua Marsh

Lying just downstream of Kawainui Marsh is 22-acre Hamakua Marsh. Cut off from Kawainui stream by a flood control levee built by the Army Corps of Engineers in 1960, Hamakua marsh became dependent on rainfall to maintain open water areas (Leone 2001). In 1995 Kaneohe Ranch donated this marsh to Ducks Unlimited, Inc who in turn gave the area to the state (Leone 2001). At this time DLNR slowly started removing invasive vegetation including Indian fleabane (*Pluchea indica),* mangrove (*Rhizophora mangle),* Christmasberry (*Schinus terebinthifolius*) and California grass (*Brachiaria mutica)* (Smith, personal communication, January 2002).

In the spring and summer of 2001, as part of a two-year, $400,000 restoration project, major vegetation removal took place to free the area of invasive mangrove (*Rhizophora mangle*) and pickleweed (*Batis maritima*) (Smith, personal communication, January 2002). Also on the agenda of the two-year project are plans to dig a fresh water well in order to supply clean water to the marsh and to place interpretive signs along Hamakua Drive (Leone, 2001). To date the only native species planted have been a'a'li'i (*Dodonaea viscosa),* milo (*Thespesia populnea),* and kou (*Cordia subcordata)* (Smith, personal communication, January 2002). However, in the spring of 2002 approximately 600 more native plants will be planted in the marsh (Smith, personal communication, January 2002). Four sedges: makaloa (*Cyperus laevigatus)*, makai (*Bulboschoenus maritimus*), 'ahu'awa (*Cyperus (Cyperus) javanicus)* and pu'uka'a (*Cyperus trachysanthos),* as well as groundcovers and shrubs such as 'akulikuli (*Sesuvium portulacastrum)*, water hyssop (*Bacopa monnieri),* and naio (*C.yoporum sandwicense)* will be planted (Smith, personal communication, January 2002). Students at Lanikai School and Kailua Elementary School are designing a "planting plan" for these native wetland plants.

Marine Corps Base Hawaii, Kaneohe Bay Hawaii

Several riparian and wetland "restoration" projects have been implemented by the Environmental Department of the Marine Corps Base on Mōkapu Peninsula in Kaneohe Bay and at Bellows Marine Corps Training Area. Projects include preserving/restoring wetlands and fishponds of Nu'upia Ponds WMA and establishment and maintenance of 3 native riparian "demonstration gardens" (Drigot 2000, Drigot 2001).

Nu'upia Ponds Wildlife Management Area encompasses 482 acres of wetland/waterbird habitat and houses approximately 10% of the state's endangered stilt (*Himantopus mexicanus knudseni*) population (Drigot 2001). Conservation projects at Nu'upia include annual removal of invasive pickleweed (*Batis maritima)* through "Mud-Ops" whereby 26-ton Amphibious Assault Vehicles are used to open up mudflats for stilt breeding and nesting (Drigot 2001).

Another major restoration project at Nu'upia Ponds WMA was the removal of red mangrove (*Rhizophora mangle*) from 20 acres of the ponds (Drigot 2000).

Three native riparian "demonstration gardens", the Muliwai and Youth Activities Center sites on the Marine Corps Base on Mokapu Peninsula and the Pūhā site at Marine Corps Training Area Bellows, have also been established by the Environmental Department of the Marine Corps Base Hawaii. Each site is located near or alongside a stream in an area with fluctuating salinity levels. Work began in February of 1999 at the Muliwai site and has continued through to the present (Diane Drigot, personal communication, November 2001). Over 1500 volunteers have helped establish these gardens. Over 3,000 plants, including a'ali'i (*Dodonaea viscosa)*, 'aka 'akai (*Schoenoplectus lacustris),* akia (*Wikstroemia uva-ursi)*, 'akoko (*Chamaesyce celastroides),* 'akulikuli (*Sesuvium portulacastrum)*, hala (*Pandanus tectorius*), 'ilima papa (*Sida fallax)*, kokio ke'oke'o (*Hibiscus waimeae)*, koki'o'ula'ula (*Hibiscus kokio*), ma'o (*Gossypium tomentosum)*, makaloa (*Cyperus laevigatus)*, loulu (*Pritchardia spp.)*, naio (*C.yoporum sandwicense)* 'ohai (*Sesbania tomentosum)*, pa'uohi'iaka (*Jacquemontia ovalifolia)*, pohinahina (*Vitex rotundifolia),* and pohuehue (*Ipomea pes-caprae)* have been planted in these "gardens" (Sustainable Resources Group, Int'l 2001). Most of the plants were selected based on availability and their ability to adapt to the environmental conditions, such as salinity and drought, encountered in the planting area (Sustainable Resources Group Int'l. 2001). Interestingly, it was also noted that once the sites were cleared of alien weeds native species such as mau'u `aki `aki (*Fimbristylis cymosa)* and kipukai (*Heliotropium curassavicum,)* which were not outplanted volunteered in at all three sites (Sustainable Resources Group Int'l. 2001).

Plants at the Muliwai site, which is adjacent to Nu'upia Ponds Wildlife Management Area, had the lowest survival rates. In fact many of the species initially planted, including all of the makaloa *(Cyperus laevigatus)*, ‘aka ‘akai (*Schoenoplectus lacustris)*, a’ali’i *(Dodonaea viscosa)* and kulu’i (*Nototrichium sandwicense),* died soon after outplanting (Sustainable Resources Group, Int’l. 2001). Makaloa (*Cyperus laevigatus)* and ‘aka ‘akai (*Schoenoplectus lacustris)* were not subsequently utilized in any of the other planting sites. By October 2001 the majority of the hala (*Pandanus tectorius)* and loulu (*Pritchardia remota*) trees were also dead and dying or showing signs of stress. The high failure rate is believed to be due to hypersaline conditions at this site (Sustainable Resources Group Int’l. 2001). Plants that have been able to survive and grow in these conditions included ‘akulikuli (*Sesuvium portulacastrum)*, naupaka (*Scaevola sericea)*, pa’uohi’iaka (*Jacquemontia ovalifolia)* and pohinahina (*Vitex rotundifolia)*. All three species required little water to become established (Sustainable Resources Group Int’l. 2001).

The plants at the Youth Activities Center (YAC) and Pūhā “gardens” showed better success and survival rates than those at the Muliwai site. Although all the kupukupu (*Nephrolepis cordifolia)* ferns planted at the YAC site and the ‘ākia (*Wikstroemia uva-ursi)* at the Pūhā sites died, the majority of plant species showed high survival rates. At YAC the naio (*C.yoporum sandwicense)*, ma’o (*Gossypium tomentosum)*, pohinahina (*Vitex rotundifolia)*, pa’uohi’iaka (*Jacquemontia ovalifolia)*, and nehe (*Lipochaeta integrifolia)* thrived even through the hot summer months (Sustainable Resources Group Int’l. 2001). At the Pūhā site the ‘akoko (*Chamaesyce celastroides)*, naio (*C.yoporum sandwicense)*, pohinahina (*Vitex rotundifolia)*, ma’o (*Gossypium tomentosum)*, ilima (*Sida fallax)* and pa’uohi’iaka (*Jacquemontia ovalifolia)* showed the fastest growth (Sustainable Resources Group Int’l 2001).

The success of these “gardens” can be attributed in part due to regular maintenance and monitoring. Irrigation and/or supplemental water appeared to be essential to survival of outplanted individuals. Irrigation, when available, decreased labor time and ensured that plants received adequate water (Sustainable Resources Group, Int'l. 2001). Constant weeding has also been necessary to keep alien weeds under control. Using herbicides prior to outplanting was found to be the best and most effective means of killing weeds, especially areas infested with grasses (Sustainable Resources Group, Int'l. 2001). Spacing plants close to one another is an additional recommendation based on outcomes at the Muliwai, YAC and Pūhā gardens (Sustainable Resources Group Int'l. 2001). Plants initially planted in dense stands tended to have fewer problems with re-colonization by alien species.

Pouhala Marsh Enhancement

Seventy-acre Pouhala Marsh is a remnant fishpond and coastal marsh located in Pearl Harbor's West Loch. This marsh is the largest remaining wetland habitat in Pearl Harbor. The USFWS has identified Pouhala Marsh as a wetland of critical concern for protection and habitat enhancement (USFWS 1995, USFWS 1998 as cited in The Natural Resource Trustees for Pearl Harbor, Oahu, Hawaii 1999). Unfortunately, of the 70 acres, 8 have been filled, 38 are overgrown, and the remaining 24 acres have been degraded through siltation and waste disposal (The Natural Resource Trustees for Pearl Harbor, Oahu, Hawaii 1999). For several years, the State of Hawaii, USFWS, Ducks Unlimited, Inc. and City and County of Honolulu have been formulating a plan for restoration of Pouhala Marsh. Although restoration has yet to begin in this marsh, project goals include clearing 20 acres of vegetation and human debris, removing 66,000 cubic yards of fill material, fencing the marsh to exclude humans and predators and creating a hydrologic link from Kapakahi Stream to the marsh (The Natural Resource Trustees for Pearl Harbor, Oahu, Hawaii 1999). Ongoing maintenance will include control of invasive flora, such as pickleweed (*Batis maritima).* Future project plans may include planting endangered Hawaiian plants to this area, but as yet no native species have been outplanted in this area.

Maui

Kealia Pond National Wildlife Refuge

Established as a National Wildlife Refuge in 1992, Kealia Pond encompasses 691 acres of wetland and dune habitat. Efforts to "restore" wetland and sand dune areas at Kealia Pond have been ongoing since approximately 1994 (Starr and Martz, personal communication, December 2001). Primary restoration activities include removal of non-native vegetation, such as pickleweed (*Batis maritima)* and Indian fleabane (*Pluchea indica),* and outplanting of native species. Outplanting efforts have largely been focused on sand dune areas of the refuge. ʻAkulikuli (*Sesuvium portulacastrum)* and ʻaki ʻaki (*Sporobolus virginicus*) have been the main species outplanted in these areas.

However, since approximately 1994, other species including ʻaweoweo (*Chenopodium oahuense),* ʻa'ali'i (*Dodonaea viscosa),* hala (*Pandanus tectorius),* ʻilima papa (*Sida fallax*), loulu (*Pritchardia spp),* ma'o (*Gossypium sandwicense*), naupaka (*Scaevola sericea)*, pa'uohi'iaka *(Jacquemontia ovalifolia),* pohinahina (*Vitex rotundifolia),* and wiliwili (*Erythrina sandwicense)* have also been planted at Kealia Pond. These outplanting efforts appear to be quite successful. When last inventoried in December 1998 the above listed species had managed to survive (Starr and Martz, personal communication, December 2001). Although only one native wetland sedge, ʻahu ʻawa (*Cyperus javanicus)* has been planted in the area, Starr and Martz (personal communication, December 2001) observed that once invasive, alien species had been removed native sedges, such as kaluha / makai (*Bulboschoenus maritimus),* were able to re-colonize cleared areas and spread from existing populations.

Kanaha Pond

Kanaha Pond is another wetland site on Maui that has benefited from continued restoration efforts. Restoration efforts at Kanaha pond began more than 10 years ago. As is typical for wetland "restoration" efforts around the state, much of the work has revolved around removal of non-native species. Approximately 20 acres of alien plants, primarily *Pluchea spp.,* koa haole (*Leucaena leucocephala),* and kiawe (*Prosopis pallida),* have been removed since 1993 (Duvall, personal communication, January 2002). Regular weeding has helped control aggressive non-native species in the area and has allowed native species to recolonize the site. Native plants that have come back solely in response to this clearing include 'aweoweo (*Chenopodium spp.)*, makaloa (*Cyperus laevigatus)*, kaluha (makai; *Bulboschoenus maritimus),* 'akulikuli (*Sesuvium portulacastrum),* water hyssop (*Bacopa monnieri)*, and pa'uohi'iaka (*Jacquemontia ovalifolia)* (Duvall, personal communication, January 2002; Native Hawaii Plant Society 1997).

Extensive planting and seeding of native plants has also been implemented at Kanaha pond. Dune / coastal species including 'akulikuli (*Sesuvium portulacastrum)* and 'aki'aki (*Sporobolus virginicus)* have been the most heavily utilized (Starr and Martz, personal communication, December 2001). In addition to planting over 500 coastal plants, volunteers have scattered seeds of makaloa (*Cyperus laevigatus)* and kaluha (makai) (*Bulboschoenus maritimus)* in the water near the edge of the pond (Native Hawaiian Plant Society 1997). Other species planted include a'ali'i (*Dodonaea viscosa),* 'akia (*Wikstroemia uva-ursi)*, 'akoko (*Chamaesyce celastroides)*, 'anaunau (*Lepidium bidentatum),* hala (*Pandanus tectorius),* naio

(*C.yoporum sandwicense)*, and wiliwili (*Erythrina sandwicensis)* (Duvall, personal communication, January 2002; Native Hawaiian Plant Society 1994).

Recently, fourteen endangered species including: *Abutilon menziesii, Cenchrus agrimoniodes, Hibiscus brackenridgei, Cyperus pinnatiformis, Pritchardia spp., Scaevola coriacea, and Sesbania tomentosa* have been planted on a trial basis to test which of these species will be able to survive at Kanaha Pond (Duvall, personal communication, January 2002). Restoration efforts will continue indefinitely at Kanaha Pond with the long-term goal of reestablishing as completely native a wetland community as possible.

‘Ahihi Kinau and Cape Hanamanioa

Anchialine ponds exist at both ‘Ahihi Kinau Natural Area Reserve and Cape Hanamanioa. These landlocked brackish ponds, which have no surface connection to the ocean but respond to tidal fluctuations via underground tubes or fissures in the lava, support many unique plants and animals. Restoration efforts at both areas have been restricted to non-native vegetation removal. Non-native species removed at ‘Ahihi Kinau include pickleweed (*Batis maritima),* Indian fleabane (*Pluchea indica*), and mangrove (*Rhizophora mangle*) (Evanson, personal communication, January 2002). Similarly, removal of non-native Indian fleabane (*Pluchea indica),* Nettleleaf goosefoot (*Chenopodium murale)* and kiawe (*Prosopis pallida*) was executed at Cape Hanamanioa (Starr and Martz, personal communication, December 2001). No outplanting has been attempted at either site.

Ulupalakua Ranch

Wetland restoration and creation has taken place at Ulupalakua Ranch as part of the Wetlands Reserve Program. This program partners the Natural Resources Conservation Service

(NRCS), Ducks Unlimited, Inc., and private landowners in efforts to protect, restore and enhance wetland habitat. In 2001 four ponds were constructed at Ulupalakua Ranch on Maui with the intent of enhancing habitat for the endangered nene goose (*Branta sandwicensis*) (Ducks Unlimited 2000, USDA-NRCS 2001). Native trees and shrubs have also been planted near the ponds.

<u>Kauai</u>

Hanalei National Wildlife Refuge

Located in the Hanalei River Valley, Hanalei NWR encompasses over 900 acres of river valley, taro farms and wooded slopes. Like other wetland wildlife refuges, Hanalei NWR was established to protect the endangered Hawaiian duck (*Anas wyvilliana*), Hawaiian moorhen (*Gallinula chloropus sandvicensis)*, Hawaiian coot (*Fulica americana alai)*, and the Hawaiian stilt (*Himantopus mexicanus knudseni)*. In 1997 ‘ahu ‘awa (*Cyperus javanicus)* was planted in several of the large ponds in Hanalei. Unfortunately, these plantings were quickly overgrown with California grass (*Brachiaria mutica)* (Asquith, personal communication, December 2001). ‘Ahu ‘awa (*Cyperus javanicus),* mamaki (*Pipturus spp.),* sawgrass (*Cladium jamaicense), Schoenoplectus juncoides,* and *Torulinium (Cyperus) oderatum* were planted in a small riparian site in 1998 (Asquith, personal communication, December 2001). Regular weeding took place for a period of a year. While the plants were able to survive while regular weeding took place no new seedlings appeared within that year. Soon after regular maintenance was halted, non-native invasive species had begun to dominate the area.

Nukoli’i

Formerly the site of an illegal sand mining operation, Nukoli'i is a tiny, slightly brackish wetland pond located near Kapa'a. In 1996 the Army Corps of Engineers and U.S. Fish and Wildlife personnel halted the sand mining operation when one Hawaiian coot (*Fulica alai*) and three common moorhens (*Gallinula chloropus sandwicensis*), including one moorhen nest, were found at the site (Environment Hawai'i 1996). Soon after the sand mining was halted, native plants including 'ae 'ae (*Bacopa monnieri)* and makai/kaluha (*Bulboschoenus maritimus)* rapidly began to re-vegetate the area where native plants had not previously existed (Kelley, personal communication, October 2001). This site is unique because it demonstrates that either viable native wetland seed banks exist and/or re-colonization by native plants is possible given the opportunity and right conditions.

Although there is no active restoration or control activities occurring, the most prominent weeds at this site, Ironwood (*Casuarina equisetifolia*) and Indian fleabane (*Pluchea indica*), do not appear to be encroaching into the pond. Non-native species perhaps have been limited by the sandy substrate of this wetland pond (Asquith, personal communication, December 2001).

Hawaii

Umikoa Ranch

Another mid to upper elevation project initiated by NRCS, Ducks Unlimited, Inc. and the Wetlands Reserve Program is located at Umikoa Ranch on the island of Hawaii. "Restoration" work at Umikoa Ranch began in 1999 and included construction of eight wetland ponds, removal of invasive vegetation and predator control (Ducks Unlimited 2000). This restoration / pond creation project is intended to benefit the endangered Hawaiian Duck or koloa (Anas wyvilliana) by providing new habitat. Although a few plants of Eleocharis calva were planted at these newly created ponds, no formal outplanting has been attempted at this site (Uyehara, personal communication, December 2001). Currently, they are attempting to recruit native plants from the seed bank by drawing the water levels down (Uyehara, personal communication, December 2001).

Kohanaiki

Located on the Kona coast, Kohanaiki consists of a series of anchialine pools and wetland ponds. In 1999 a youth project was undertaken to help "restore" these wetlands and anchialine ponds. Activities included: surveying the flora and fauna of the area, removal of weeds such as pickleweed (*Batis maritima*) and kiawe (*Prosopis pallida)*, and establishment of a solar powered, brackish water irrigation system intended for propagation of native plants (Kohanaiki 'Ohana 2001, http://www.kohanaiki.org/). Although some native plants have been outplanted at this site, primary efforts have focused on removal of invasive alien species and encouraging the growth of native plants, such as makaloa (*Cyperus laevigatus)*, which is already growing at this site.

Molokai

Kakahaia National Wildlife Refuge

Kakahaia National Wildlife Refuge was established in 1977 and contains a 15-acre spring-fed pond and a seven-acre manmade impoundment. Surrounding two sides of the old pond are thick kiawe (*Prosopis pallida)* trees. Several years ago non-native vegetation, such as Indian fleabane (*Pluchea indica),* was treated with herbicide and many of the large kiawe (*Prosopis pallida)* trees in the mudflats adjacent to the ponds were removed (Starr and Martz, personal communication, January 2001). At this time ‘akulikuli (*Sesuvium portulacastrum)* was also planted and is still flourishing (Starr and Martz, personal communication, January 2001).

DISCUSSION

As this review demonstrates, wetland “restoration” in Hawai’i has primarily focused on removal of non-native vegetation. Re-vegetating an area with native species is rarely attempted. Despite the lack of outplanting efforts, encouraging results have been achieved in many areas. Native plants have been found to spread and colonize in response just to removal of non-native vegetation. Unfortunately, this has not been monitored or documented and it is unknown whether these native species are able to persist. Presumably, control of invasive species will need to be continued in order to maintain these populations of native species.

Lessons have also been learned and recommendations can be offered from projects where outplanting of native plants has been attempted. Supplemental water and persistent maintenance seem to be common keys to native re-vegetation efforts (Sustainable Resources Group Int’l, Inc. 2001; Starr and Martz, personal communication December 2001). Initial planting of native species in dense stands may also be important. Dense plantings may help limit recruitment of non-native weeds, and although more expensive initially, in the long run this strategy can save money in maintenance costs (Sustainable Resources Group Int’l. 2001). Further restoration efforts would benefit greatly from documentation and continued monitoring to assess the success of restoration and particularly outplanting efforts.

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